CHALMERS UNIVERSITY OF TECHNOLOGY

Examination in Bioinformatics, MVE360

Monday 11 March 2013, 08:30-12:30

Examiner: Graham Kemp (telephone 772 54 11, room 6475 EDIT)
The examiner will visit the exam room at 09:30 and 11:30.

Results: Will be published by 26 March 2013 at the latest.

Exam review: See course web page for time and place:
http://www.cse.chalmers.se/edu/year/2013/course/MVE360/

Grades: Grades for Chalmers students are normally determined as follows:
$\geq$ 48 for grade 5; $\geq$ 36 for grade 4; $\geq$ 24 for grade 3.

Grades for GU students are normally determined as follows:
$\geq$ 42 for grade VG; $\geq$ 24 for grade G.

Help material: None
English language dictionaries are allowed.

Specific instructions:

- Check that you have received:
  - question paper (5 pages)
  - EMBL nucleotide sequence database entry SCYSP3G (1 page)
  - Perl reference guide (4 pages)
  - HMM formula collection (3 pages)
- Please answer in English where possible. You may clarify your answers in Swedish if you are not confident you have expressed yourself correctly in English.
- Begin the answer to each question on a new page.
- Write clearly; unreadable = wrong!
- Fewer points are given for unnecessarily complicated solutions.
- Indicate clearly if you make any assumptions that are not given in the question.
- Write the page number and question number on every page.

Question 1. What output is printed when the following program is run?
4 p

```
#!/usr/bin/perl -w

$s = "atccatattt"; if ( $s =~ /c(.*)$/ )        { print "a) $1\n"; }
$s = "atccatattt"; if ( $s =~ /(a+)t/ )         { print "b) $1\n"; }
$s = "atccatattt"; if ( $s =~ /.*(..)a(.*)..$/ ) { print "c) $1, $2\n"; }
$s = "atccatattt"; if ( $s =~ /(.)\1*$/ )       { print "d) $1\n"; }

$s = "atccatattt"; $s =~ s/a./g/g    ; print "e) $s\n";
$s = "atccatattt"; $s =~ s/.*at//g   ; print "f) $s\n";
$s = "atccatattt"; $s =~ s/t.*t/g/   ; print "g) $s\n";
$s = "atccatattt"; $s =~ tr/ta/cg/   ; print "h) $s\n";
```

(4p)

Question 2. 6 p

a) Draw a suffix tree for the string "ATCATC".
(3p)

b) Write a Perl program that prompts the user to type in a string, reads that string, and then prints out all suffixes of that string.
(3p)

Question 3. In an EMBL nucleotide sequence database entry, the positions where the coding sequence starts and ends are given on the feature table line (beginning "FT") for feature "CDS" (e.g. positions 908 and 2293 in EMBL entry SCYSP3G, see attached sheet).
12 p

a) Write a Perl program that reads the start and end positions of the coding sequence from an EMBL database file whose name is specified on the command line. The program should read the full sequence, and store the coding sequence in the variable $cs.
(4p)

b) Extend your solution to part (a) so that the program writes the coding sequence to standard output, with three nucleotides (one codon) per line.
(3p)

c) The codons "aca", "acc", "acg" and "act" all code for the amino acid threonine. Extend your solution to part (b) so that the program counts how often each of these four codons occurs in the coding sequence, and writes out the threonine codon(s) that occurs most often.
(5p)

Question 4. 4 p

Using a gap score of -2 and match/mismatch scores taken from the PAM250 substitution matrix (given below), derive the score matrix for a global alignment of “QFN” with “NGYE”.

In this case, what is the score of an optimal global alignment? Give the alignment(s) with this score.

PAM250 substitution matrix:

	A	R	N	D	C	Q	E	G	H	I	L	K	M	F	P	S	T	W	Y	V
A	2																			
R	-2	6																		
N	0	0	2																	
D	0	-1	2	4																
C	-2	-4	-4	-5	4															
Q	0	1	1	2	-5	4														
E	0	-1	1	3	-5	2	4													
G	1	-3	0	1	-3	-1	0	5												
H	-1	2	2	1	-3	3	1	-2	6											
I	-1	-2	-2	-2	-2	-2	-2	-3	-2	5										
L	-2	-3	-3	-4	-6	-2	-3	-4	-2	2	6									
K	-1	3	1	0	-5	1	0	-2	0	-2	-3	5								
M	-1	0	-2	-3	-5	-1	-2	-3	-2	2	4	0	6							
F	-4	-4	-4	-6	-4	-5	-5	-5	-2	1	2	-5	0	9						
P	1	0	-1	-1	-3	0	-1	-1	0	-2	-3	-1	-2	-5	6					
S	1	0	1	0	0	-1	0	1	-1	-1	-3	0	-2	-3	1	3				
T	1	-1	0	0	-2	-1	0	0	-1	0	-2	0	-1	-2	0	1	3			
W	-6	2	-4	-7	-8	-5	-7	-7	-3	-5	-2	-3	-4	0	-6	-2	-5	17		
Y	-3	-4	-2	-4	0	-4	-4	-5	0	-1	-1	-4	-2	7	-5	-3	-3	0	10	
V	0	-2	-2	-2	-2	-2	-2	-1	-2	4	2	-2	2	-1	-1	-1	0	-6	-2	4

(4p)

Question 5. 4 p

a) Draw all possible *rooted* trees with four different external nodes.
(2p)

b) In the context of *maximum parsimony*, what is meant by an *informative site*? In the simple multiple alignment below, indicate the sites (columns in the alignment) that are informative.

```
GCTGT
GCTGG
ACACG
ACAGG
```

(2p)

Question 6. 6 p

a) Explain the property that characterizes a Markov Chain and make it a special case of a stochastic process.

(1p)

b) Explain the *processes* that compound a HMM. Which *parameters* describe the model? What is the difference between a HMM and a Markov Chain?

(3p)

c) Once you have trained a HMM, i.e. found the parameters, which algorithm would you use to parse a new DNA sequence? Briefly explain.

(1p)

d) How does a pair HMM differ from a standard HMM?

(1p)

Question 7. 4 p

Genes have variants called alleles. Each person has 2 copies (alleles) of a gene. Consider one gene with two alleles A and a in a population. So, in our supposed population we can have genotypes (possible combination of alleles for the same gene) $1 = AA$, $2 = aa$ and $3 = Aa$ with probabilities p_1, p_2 and p_3, respectively. We will consider our population is in the so called Hardy-Weinberg equilibrium, meaning that the genotype proportions remain the same from one generation to the next. For simplicity we will also consider that each individual has only one offspring. Now, starting with a randomly chosen individual in the population, at some point in time (generation 1), we let X_n = the genotype of her descendant in the nth generation.

a) Explain why $\{X_n, n = 1, 2, ...\}$ is a Markov Chain.

(1p)

b) What are the missing transitions, a_{13} and a_{22} in the transition matrix below?

$$\begin{bmatrix} p_1 + \frac{p_3}{2} & 0 & ? \\ 0 & ? & p_1 + \frac{p_3}{2} \\ \frac{p_1}{2} + \frac{p_3}{4} & \frac{p_2}{2} + \frac{p_3}{4} & \frac{p_1}{2} + \frac{p_2}{2} + \frac{p_3}{2} \end{bmatrix}$$

(2p)

c) If an individual has genotype Aa, what is the probability that her grandchild will have genotype Aa as well? Show how this probability can be calculated from the transition probabilities.

(1p)

Question 8. 4 p

Assume that we have a very simple HMM model that classifies stretches of genes in DNA sequences as exons (coding) or introns (non-coding). The emission probabilities are given in below:

	P(A)	P(C)	P(G)	P(T)
E	0.25	0.25	0.25	0.25
I	0.4	0.1	0.1	0.4

The transition probabilities between different states are 0.1, i.e
$a_{IE} = a_{EI} = 0.1$.
States are initiated with the same probability.

Find the optimal state sequence for the DNA stretch $TAAG$.

(4p)

Question 9. 2 p

Using a pair HMM it is possible to calculate the a score of similarity for two sequences Y_1^T and Z_1^U (where $Y_1^T = Y_1, ..., Y_T$ and $Z_1^T = Z_1, ..., Z_T$):

$$log \frac{P(Y_1^T, Z_1^U)}{P(Y_1^T, Z_1^U|R)} \tag{1}$$

The probability in the numerator is calculated under the assumption that the two sequences are generated by by the pair HMM, and the probability in the denominator is calculated under the assumption that they are randomly assembled, independently of each other.

a) By which algorithm can $P(Y_1^T, Z_1^U)$ be determined?
(1p)

b) In the context of sequence alignment, how is $P(Y_1^T, Z_1^U)$ to be interpreted?
(1p)

Question 10. 14 p

a) In metagenomics one sometimes talks about a *gene-centric analysis* approach. Describe what this means. (2-5 sentences)
(4p)

b) Describe and compare (giving advantages and disadvantages) two of the next generation sequencing techniques. Computational aspects should be included in your discussion.
(6p)

c) What is a *paired-end* sequencing technique, and how does a paired-end approach help in *de novo* genome assembly?
(4p)

EMBL Nucleotide Sequence Database Entry SCYSP3G

```
ID   SCYSP3G    standard; genomic DNA; FUN; 2500 BP.
XX
AC   M77197;
XX
SV   M77197.1
XX
DT   09-OCT-1991 (Rel. 29, Created)
DT   17-APR-2005 (Rel. 83, Last updated, Version 3)
XX
DE   Yeast subtilisin-like protease III gene, complete cds.
XX
KW   subtilisin-like protease III.
XX
OS   Saccharomyces cerevisiae (baker’s yeast)
OC   Eukaryota; Fungi; Ascomycota; Saccharomycotina; Saccharomycetes;
OC   Saccharomycetales; Saccharomycetaceae; Saccharomyces.
XX
RN   [1]
RP   1-2500
RA   Mason O.B., Wong P.A., Barr P.J.;
RT   "Nucleotide sequence of YSP3, a new subtilisin-like protease from
RT   Saccharomyces cerevisiae";
RL   Unpublished.
XX
FH   Key             Location/Qualifiers
FH
FT   source          1..2500
FT                   /db_xref="taxon:4932"
FT                   /mol_type="genomic DNA"
FT                   /organism="Saccharomyces cerevisiae"
FT                   /strain="S288C"
FT   sig_peptide     908..958
FT                   /product="subtilisin-like protease III"
FT                   /gene="YSP3"
FT   CDS             908..2293
FT                   /codon_start=1
FT                   /db_xref="GOA:P25036"
FT                   /db_xref="HSSP:1DBI"
FT                   /db_xref="UniProtKB/Swiss-Prot:P25036"
FT                   /product="subtilisin-like protease III"
FT                   /gene="YSP3"
FT                   /protein_id="AAA35237.1"
FT                   /translation="MKFSTILPILWANCCLCMIIPEFDGIVRFIENIDGTRSVRAGEGL
FT                   GQHDPGNFHTEHQHVAHKTEFLPYRYVIVFNEDISLQQIQSHMQVVQKDHSTSVGKLTE
FT                   DDAFWRVISSSVSSKSQFGGIDNFFDINGLFRGYTGYFTDEIIKIISQDPIIKFVEQET
FT                   TVKISNSSLQEEAPWGLHRVSHREKGVTSYVLDTGIDTEHEDFEGRAEWGAVIPANDEA
FT                   SDLNGHGTHCAGIIGSKHFGVAKNTKIVAVKVLRSNGEGTVSDVIKGIELVTKEHIESS
FT                   KKKNKEFKGSTANLSLGSSKSLAMEMAVNAAVDSGVHFAIAAGNEDEDACLSSPAGAEK
FT                   SITVGASTFSDDRAFFSNWGTCVDVFAPGINIMSTYIGSRNATLSLSGTSMASPHVAGI
FT                   LSYFLSLQPAPDSEFFNDAPSPQELKEKVLKFSTQGVLGDIGDDTPNKLIYNGGGKKLD
FT                   GFW"
FT   mat_peptide     959..2290
FT                   /product="subtilisin-like protease III"
FT                   /gene="YSP3"
XX
SQ   Sequence 2500 BP; 736 A; 436 C; 516 G; 812 T; 0 other;
     atgagaaaac tatcctcgtc ccacttttgc ctattacact actctactcc tctactgatt        60
     ggaatgttct atctcttgta agttggataa acaatgtggc tttgtttacg ctatggcctt       120
     tgttgaaaaa ggacggtctt catttacagt atgccgtatc tttcttacta agcaattggc       180
     tgattggaaa tttcagtttt attacaccaa ggttcttgcc aaaatcttta actcctggcc       240
     cttctatcag cagcatcaat agcgactata gaagaagaag cttactgcca tataatgtgg       300
     tttggaaaag ttttatcata ggaacgtata ttgctatggg cttttatcat ttcttagatc       360
     aatttgtagc acctccatcg aaatatccag acttgtgggt gttgttgaac tgtgctgttg       420
     ggttcatttg ctttagcata ttttggctat ggtcttatta caagatattc acttccggta       480
     gcaaatccat gttggacttg tagttacatt ttaacttata taactgtaca atgtcagtca       540
     gacgcaactg atataaaaga tgtgctctac tagtaaagat tatttcaact cttaaaaata       600
     tttgcaaaaa tgcactataa tttgatctaa ataaggtaca gataatggta aggcaaaact       660
     actacatttg gcagtgagta ttgttagctt ttccgcttat tttctctcct ggagcagtca       720
     ccgttcgcgt cttccgagca atggcgaatc tcgaatctcg gaagggcggc tactgataac       780
     aggcactcat atatatatta ttagataaga gttttttttg actgagtcct tataacacgt       840
     aacttatcaa gttgctatat tataagttca ggtttccaat ctgttcaacc actttatatc       900
     tttaaacatg aaattctcta ccatcttacc tatactttgg gcaaattgct gtctttgtat       960
     gatcatccct gaattcgacg ggatagttcg ttttattgag aatattgatg gtactcgcag      1020
     tgtaagagca ggagaaggtt tgggacagca tgatcctgga aattttcaca ctgaacatca      1080
     acatgtcgct cataaaacag agtttctacc ttatcggtat gtcatagtct ttaatgaaga      1140
     tatttccctc cagcagattc aatcgcatat gcaagtggta cagaaggatc atagtacctc      1200
     agtaggtaag cttacagaag atgacgcttt ttggagggta atttcttctt cagtatcatc      1260
     taaatctcaa tttggaggta ttgataattt ttttgacata aatggtctat ttcgaggtta      1320
     tactggctat ttcactgatg aaatcatcaa aataatctcc caggacccaa tcataaagtt      1380
     cgtagagcaa gaaactacag taaaaatatc taattcgtca ttgcaggaag aagcgccttg      1440
     gggtttacat agagtttcgc acagggaaaa aggtgtcaca tcatatgtac tcgacactgg      1500
     aattgatacc gaacacgagg actttgaagg gcgtgctgag tggggagccg ttataccagc      1560
     aaacgatgaa gcttctgatt tgaatggtca tggaactcac tgtgcgggga ttatcggatc      1620
     gaagcacttt ggtgtagcca aaaatacaaa aatagtagct gtcaaagttc ttcgttctaa      1680
     tggggagggg acggtttcag atgttattaa aggtatagag ctcgttacta aggagcatat      1740
     agaatcgtcg aagaaaaaga ataaagaatt caaaggatca actgccaatc tttctttggg      1800
     aagtagcaaa tcgctagcta tggaaatggc tgtaaatgca gctgtagata gcggtgtcca      1860
     ttttgctatt gcggcaggaa atgaggatga agatgcttgt ctcagttcac cagcaggagc      1920
     tgaaaaaagt attacggtcg gtgcttcgac atttagtgat gatagagcat tcttttcgaa      1980
     ctggggcaca tgtgttgatg tgtttgctcc cggtataaat attatgtcca cctatattgg      2040
     ttcaaggaat gcaactttaa gtttatcagg tacttccatg gcgtccccgc acgttgctgg      2100
     tattttaagt tactttttat cattacagcc tgcaccagac agtgaatttt tcaacgacgc      2160
     tccctcacct caagaattga aagaaaaagt cctaaaattt agtacacagg gagtactggg      2220
     tgatatcggt gatgatactc ctaacaaatt aatatataat gggggaggta agaaattgga      2280
     tggtttttgg tagctttgat cttgaattta ggagcactca ggaggtttta ttttattctt      2340
     ttcatacagg tcagcataag cctaagaaag acgtttgaaa tcgctaatcg gctttttgtt      2400
     cctacgagta ggttaaattc tttcaataat atacataata aattagtagt aaagataaac      2460
     ttttgttcgc gatgcagttc ttttggttgt gaatagtttt                              2500
//
```

Perl 5 Reference Guide (Extract)

Literals

Numeric:
123
1_234
123.4
5E-10
0xff (hex)
0377 (octal)

String:
'abc'
literal string, no variable interpolation or escape characters, except \' and \\. Also: q/abc/.
Almost any pair of delimiters can be used instead of /.../.
"abc"
Variables are interpolated and escape sequences are processed. Also: qq/abc/.
Escape sequences: \t (Tab), \n (Newline), \r (Return), \f (Formfeed), \b (Backspace), \a (Alarm), \e (Escape), \033 (octal), \x1b (hex), \c[(control)
\l and \u lowercase/uppercase the following character. \L and \U lowercase/uppercase until a \E is encountered. \Q quote regular expression characters until a \E is encountered.
`COMMAND`
evaluates to the output of the COMMAND. Also: qx/COMMAND/.

Array:
(1, 2, 3). () is an empty array.
(1..4) is the same as (1,2,3,4),
likewise ('a'..'z').
qw/foo bar .../ is the same as ('foo','bar',...).

Array reference:
[1,2,3]

Hash (associative array):
(KEY1, VAL1, KEY2, VAL2,...)
Also (KEY1=> VAL1, KEY2=> VAL2,...)

Hash reference:
{KEY1, VAL1, KEY2, VAL2,...}

Code reference:
sub {STATEMENTS}

Filehandles:
<STDIN>, <STDOUT>, <STDERR>, <ARGV>, <DATA>.
User-specified: HANDLE, $VAR.

Globs:
<PATTERN> evaluates to all filenames according to the pattern. Use <${VAR}> or glob $VAR to glob from a variable.

Here-Is:
<<IDENTIFIER
Shell-style "here document."

Special tokens:
__FILE__: filename; __LINE__: line number; __END__: end of program; remaining lines can be read using the filehandle DATA.

Variables

$var	a simple scalar variable.
$var[28]	29th element of array @var.
$p = \@var	now $p is a reference to array @var.
$$p[28]	29th element of array referenced by $p. Also, $p->[28].
$var[-1]	last element of array @var.
$var[$i][$j]	$jth element of the $ith element of array @var.
$var{'Feb'}	one value from hash (associative array) %var.
$p = \%var	now $p is a reference to hash %var.
$$p{'Feb'}	a value from hash referenced by $p. Also, $p->{'Feb'}.
$#var	last index of array @var.
@var	the entire array; in a scalar context, the number of elements in the array.
@var[3,4,5]	a slice of array @var.
@var{'a','b'}	a slice of %var; same as ($var{'a'},$var{'b'}).
%var	the entire hash; in a scalar context, true if the hash has elements.
$var{'a',1,...}	emulates a multidimensional array.
('a'...'z')[4,7,9]	a slice of an array literal.
PKG::VAR	a variable from a package, e.g., $pkg::var, @pkg::ary.
\OBJECT	reference to an object, e.g., \$var, \%hash.
*NAME	refers to all objects represented by NAME. *n1 = *n2 makes n1 an alias for n2. *n1 = $n2 makes $n1 an alias for $n2.

You can always use a {BLOCK} returning the right type of reference instead of the variable identifier, e.g., ${...}, &{...}. $$p is just a shorthand for ${$p}.

Operators

**	Exponentiation
+ - * /	Addition, subtraction, multiplication, division
%	Modulo division
& | ^	Bitwise AND, bitwise OR, bitwise exclusive OR
>> <<	Bitwise shift right, bitwise shift left
|| &&	Logical OR, logical AND
.	Concatenation of two strings
x	Returns a string or array consisting of the left operand (an array or a string) repeated the number of times specified by the right operand

All of the above operators also have an assignment operator, e.g., .=

->	Dereference operator
\	Reference (unary)
! ~	Negation (unary), bitwise complement (unary)
++ --	Auto-increment (magical on strings), auto-decrement
== !=	Numeric equality, inequality
eq ne	String equality, inequality
< >	Numeric less than, greater than
lt gt	String less than, greater than
<= >=	Numeric less (greater) than or equal to
le ge	String less (greater) than or equal to
<=> cmp	Numeric (string) compare. Returns -1, 0, 1.
=~ !~	Search pattern, substitution, or translation (negated)
..	Range (scalar context) or enumeration (array context)
?:	Alternation (if-then-else) operator
,	Comma operator, also list element separator. You can also use =>.
not	Low-precedence negation
and	Low-precedence AND
or xor	Low-precedence OR, exclusive OR

All Perl functions can be used as list operators, in which case they have very high or very low precedence, depending on whether you look at the left or the right side of the operator. Only the operators **not**, **and**, **or** and **xor** have lower precedence.

A "list" is a list of expressions, variables, or lists. An array variable or an array slice may always be used instead of a list.

Parentheses can be added around the parameter lists to avoid precedence problems.

Statements

Every statement is an expression, optionally followed by a modifier, and terminated by a semicolon. The semicolon may be omitted if the statement is the final one in a BLOCK.

Execution of expressions can depend on other expressions using one of the modifiers **if**, **unless**, **while** or **until**, for example:

> EXPR1 **if** EXPR2 **;**
> EXPR1 **until** EXPR2 **;**

The logical operators `||`, `&&` or `?:` also allow conditional execution:

> EXPR1 **||** EXPR2 **;**
> EXPR1 **?** EXPR2 **:** EXPR3 **;**

Statements can be combined to form a BLOCK when enclosed in `{}`. Blocks may be used to control flow:

> **if (**EXPR**)** BLOCK [[**elsif (**EXPR**)** BLOCK ...] **else** BLOCK]
> **unless (**EXPR**)** BLOCK [**else** BLOCK]
> [LABEL**:**] **while** (EXPR) BLOCK [**continue** BLOCK]
> [LABEL**:**] **until** (EXPR) BLOCK [**continue** BLOCK]
> [LABEL**:**] **for** (EXPR**;** EXPR**;** EXPR) BLOCK
> [LABEL**:**] **foreach** VAR[2] (LIST) BLOCK
> [LABEL**:**] BLOCK [**continue** BLOCK]

Program flow can be controlled with:

goto LABEL	Continue execution at the specified label.
last [LABEL]	Immediately exits the loop in question. Skips continue block.
next [LABEL]	Starts the next iteration of the loop.
redo [LABEL]	Restarts the loop block without evaluating the conditional again.

Special forms are:

> **do** BLOCK **while** EXPR **;**
> **do** BLOCK **until** EXPR **;**

which are guaranteed to perform BLOCK once before testing EXPR, and

> **do** BLOCK

which effectively turns BLOCK into an expression.

Structure Conversion

pack TEMPLATE**,** LIST
: Packs the values into a binary structure using TEMPLATE.

unpack TEMPLATE**,** EXPR
: Unpacks the structure EXPR into an array, using TEMPLATE.

TEMPLATE is a sequence of characters as follows:

a / **A**	ASCII string, null- / space-padded
b / **B**	Bit string in ascending / descending order
c / **C**	Native / unsigned char value
f / **d**	Single / double float in native format
h / **H**	Hex string, low / high nybble first
i / **I**	Signed / unsigned integer value
l / **L**	Signed / unsigned long value
n / **N**	Short / long in network (big endian) byte order
s / **S**	Signed / unsigned short value
u / **P**	Uuencoded string / pointer to a string
v / **V**	Short / long in VAX (little endian) byte order
x / **@**	Null byte / null fill until position
X	Backup a byte

Each character may be followed by a decimal number that will be used as a repeat count; an asterisk (`*`) specifies all remaining arguments. If the format is preceded with `%`N, **unpack** returns an N-bit checksum instead. Spaces may be included in the template for readability purposes.

String Functions

chomp LIST[2]
: Removes line endings from all elements of the list; returns the (total) number of characters removed.

chop LIST[2]
: Chops off the last character on all elements of the list; returns the last chopped character.

crypt PLAINTEXT**,** SALT
: Encrypts a string.

eval EXPR[2]
: EXPR is parsed and executed as if it were a Perl program. The value returned is the value of the last expression evaluated. If there is a syntax error or runtime error, an undefined string is returned by **eval**, and `$@` is set to the error message. See also **eval** in section Miscellaneous.

index STR**,** SUBSTR [**,** OFFSET]
: Returns the position of SUBSTR in STR at or after OFFSET. If the substring is not found, returns `-1` (but see `$[` in section Special Variables).

length EXPR[2]
: Returns the length in characters of the value of EXPR.

lc EXPR
: Returns a lowercase version of EXPR.

lcfirst EXPR
: Returns EXPR with the first character lowercase.

quotemeta EXPR
: Returns EXPR with all regular expression metacharacters quoted.

rindex STR**,** SUBSTR [**,** OFFSET]
: Returns the position of the last SUBSTR in STR at or before OFFSET.

substr EXPR**,** OFFSET [**,** LEN]
: Extracts a substring of length LEN out of EXPR and returns it. If OFFSET is negative, counts from the end of the string. May be assigned to.

uc EXPR
: Returns an uppercased version of EXPR.

ucfirst EXPR
: Returns EXPR with the first character uppercased.

Array and List Functions

delete `$`HASH`{`KEY`}`
: Deletes the specified value from the specified hash. Returns the deleted value unless HASH is **tie**d to a package that does not support this.

each `%`HASH
: Returns a 2-element array consisting of the key and value for the next value of the hash. Entries are returned in an apparently random order. After all values of the hash have been returned, a null array is returned. The next call to **each** after that will start iterating again.

exists EXPR[2]
: Checks if the specified hash key exists in its hash array.

grep EXPR**,** LIST
grep BLOCK LIST
: Evaluates EXPR or BLOCK for each element of the LIST, locally setting `$_` to refer to the element. Modifying `$_` will modify the corresponding element from LIST. Returns the array of elements from LIST for which EXPR returned **true**.

join EXPR**,** LIST
: Joins the separate strings of LIST into a single string with fields separated by the value of EXPR, and returns the string.

keys `%`HASH
: Returns an array with all of the keys of the named hash.

map EXPR**,** LIST
map BLOCK LIST
: Evaluates EXPR or BLOCK for each element of the LIST, locally setting `$_` to refer to the element. Modifying `$_` will modify the corresponding element from LIST. Returns the list of results.

pop `@`ARRAY
: Pops off and returns the last value of the array.

push `@`ARRAY**,** LIST

Pushes the values of LIST onto the end of the array.

reverse LIST
: In array context, returns the LIST in reverse order. In scalar context: returns the first element of LIST with bytes reversed.

scalar @ARRAY
: Returns the number of elements in the array.

scalar %HASH
: Returns a **true** value if the hash has elements defined.

shift [@ARRAY]
: Shifts the first value of the array off and returns it, shortening the array by 1 and moving everything down. If @ARRAY is omitted, shifts **@ARGV** in main and **@_** in subroutines.

sort [SUBROUTINE] LIST
: Sorts the LIST and returns the sorted array value. SUBROUTINE, if specified, must return less than zero, zero, or greater than zero, depending on how the elements of the array (available to the routine as **$a** and **$b**) are to be ordered. SUBROUTINE may be the name of a user-defined routine, or a BLOCK.

splice @ARRAY**,** OFFSET [**,** LENGTH [**,** LIST]]
: Removes the elements of @ARRAY designated by OFFSET and LENGTH, and replaces them with LIST (if specified). Returns the elements removed.

split [PATTERN [**,** EXPR[2] [**,** LIMIT]]]
: Splits a string into an array of strings, and returns it. If LIMIT is specified, splits into at most that number of fields. If PATTERN is also omitted, splits at the whitespace. If not in array context, returns number of fields and splits to **@_**. See also Search and Replace Functions.

unshift @ARRAY**,** LIST
: Prepends list to the front of the array, and returns the number of elements in the new array.

values %HASH
: Returns a normal array consisting of all the values of the named hash.

Regular Expressions

Each character matches itself, unless it is one of the special characters **+ ? . * ^ $ ( ) [ ] { } | **. The special meaning of these characters can be escaped using a ****.

. matches an arbitrary character, but not a newline unless it is a single-line match (see **m//s**).
(...**)** groups a series of pattern elements to a single element.
^ matches the beginning of the target. In multiline mode (see **m//m**) also matches after every newline character.
$ matches the end of the line. In multiline mode also matches before every newline character.
[... **]** denotes a class of characters to match. **[^** ... **]** negates the class.
(... **|** ... **|** ... **)** matches one of the alternatives.
(?# TEXT **)** Comment.
(?: REGEXP **)** Like **(**REGEXP**)** but does not make back-references.
(?= REGEXP **)** Zero width positive look-ahead assertion.
(?! REGEXP **)** Zero width negative look-ahead assertion.
(? MODIFIER **)** Embedded pattern-match modifier. MODIFIER can be one or more of **i**, **m**, **s**, or **x**.

Quantified subpatterns match as many times as possible. When followed with a **?** they match the minimum number of times. These are the quantifiers:

+ matches the preceding pattern element one or more times.
? matches zero or one times.
***** matches zero or more times.
{N**,**M**}** denotes the minimum N and maximum M match count. **{**N**}** means exactly N times; **{**N**,}** means at least N times.

A **** escapes any special meaning of the following character if non-alphanumeric, but it turns most alphanumeric characters into something special:

\w matches alphanumeric, including _, **\W** matches non-alphanumeric.
\s matches whitespace, **\S** matches non-whitespace.
\d matches numeric, **\D** matches non-numeric.
\A matches the beginning of the string, **\Z** matches the end.
\b matches word boundaries, **\B** matches non-boundaries.
\G matches where the previous **m//g** search left off.
\n, **\r**, **\f**, **\t** etc. have their usual meaning.

\w, **\s** and **\d** may be used within character classes, **\b** denotes backspace in this context.

Back-references:

\1 ... **\9** refer to matched subexpressions, grouped with **()**, inside the match.
\10 and up can also be used if the pattern matches that many subexpressions.

See also **$1** ... **$9**, **$+**, **$&**, **$'**, and **$'** in section Special Variables.

With modifier **x**, whitespace can be used in the patterns for readability purposes.

Search and Replace Functions

[EXPR **=~**] [**m**] **/**PATTERN**/** [**g**] [**i**] [**m**] [**o**] [**s**] [**x**]
: Searches EXPR (default: **$_**) for a pattern. If you prepend an **m** you can use almost any pair of delimiters instead of the slashes. If used in array context, an array is returned consisting of the subexpressions matched by the parentheses in the pattern, i.e., **($1,$2,$3,**...**)**.
: Optional modifiers: **g** matches as many times as possible; **i** searches in a case-insensitive manner; **o** interpolates variables only once. **m** treats the string as multiple lines; **s** treats the string as a single line; **x** allows for regular expression extensions.
: If PATTERN is empty, the most recent pattern from a previous match or replacement is used.
: With **g** the match can be used as an iterator in scalar context.

?PATTERN**?**
: This is just like the **/**PATTERN**/** search, except that it matches only once between calls to the **reset** operator.

[**$**VAR **=~**] **s/**PATTERN**/**REPLACEMENT**/** [**e**] [**g**] [**i**] [**m**] [**o**] [**s**] [**x**]
: Searches a string for a pattern, and if found, replaces that pattern with the replacement text. It returns the number of substitutions made, if any; if no substitutions are made, it returns **false**.
: Optional modifiers: **g** replaces all occurrences of the pattern; **e** evaluates the replacement string as a Perl expression; for any other modifiers, see **/**PATTERN**/** matching. Almost any delimiter may replace the slashes; if single quotes are used, no interpretation is done on the strings between the delimiters, otherwise the strings are interpolated as if inside double quotes.
: If bracketing delimiters are used, PATTERN and REPLACEMENT may have their own delimiters, e.g., **s(foo)[bar]**. If PATTERN is empty, the most recent pattern from a previous match or replacement is used.

[**$**VAR **=~**] **tr/**SEARCHLIST**/**REPLACEMENTLIST**/** [**c**] [**d**] [**s**]
: Translates all occurrences of the characters found in the search list with the corresponding character in the replacement list. It returns the number of characters replaced. **y** may be used instead of **tr**.
: Optional modifiers: **c** complements the SEARCHLIST; **d** deletes all characters found in SEARCHLIST that do not have a corresponding character in REPLACEMENTLIST; **s** squeezes all sequences of characters that are translated into the same target character into one occurrence of this character.

pos SCALAR
: Returns the position where the last **m//g** search left off for SCALAR. May be assigned to.

study [**$**VAR[2]]
: Study the scalar variable **$**VAR in anticipation of performing many pattern matches on its contents before the variable is next modified.

Input / Output

In input/output operations, FILEHANDLE may be a filehandle as opened by the **open** operator, a predefined filehandle (e.g., **STDOUT**) or a scalar variable that evaluates to the name of a filehandle to be used.

<FILEHANDLE**>**
: In scalar context, reads a single line from the file opened on FILEHANDLE. In array context, reads the whole file.

< >
: Reads from the input stream formed by the files specified in **@ARGV**, or standard input if no arguments were supplied.

close FILEHANDLE
: Closes the file or pipe associated with the filehandle.

open FILEHANDLE [**,** FILENAME]
: Opens a file and associates it with FILEHANDLE. If FILENAME is omitted, the scalar variable of the same name as the FILEHANDLE must contain the filename.
: The following filename conventions apply when opening a file.

`"FILE"` open FILE for input. Also `"<FILE"`.
`">FILE"` open FILE for output, creating it if necessary.
`">>FILE"` open FILE in append mode.
`"+<FILE"` open FILE with read/write access (file must exist).
`"+>FILE"` open FILE with read/write access (file truncated).
`"|CMD"` opens a pipe to command CMD; forks if CMD is `-`.
`"CMD|"` opens a pipe from command CMD; forks if CMD is `-`.
FILE may be `&`FILEHND in which case the new filehandle is connected to the (previously opened) filehandle FILEHND. If it is `&=`N, FILE will be connected to the given file descriptor. **open** returns **undef** upon failure, **true** otherwise.

Returns a pair of connected pipes.

print [FILEHANDLE] [LIST²]
Equivalent to **print** FILEHANDLE **sprintf** LIST.

Special Variables

The following variables are global and should be localized in subroutines:

`$_` The default input and pattern-searching space.
`$.` The current input line number of the last filehandle that was read.
`$/` The input record separator, newline by default. May be multicharacter.
`$,` The output field separator for the print operator.
`$"` The separator that joins elements of arrays interpolated in strings.
`$\` The output record separator for the print operator.
`$#` The output format for printed numbers. Deprecated.
`$*` Set to 1 to do multiline matching within strings. Deprecated, see the **m** and **s** modifiers in section Search and Replace Functions.
`$?` The status returned by the last `‘...‘` command, pipe **close** or **system** operator.
`$]` The perl version number, e.g., `5.001`.
`$[` The index of the first element in an array, and of the first character in a substring. Default is 0. Deprecated.
`$;` The subscript separator for multidimensional array emulation. Default is "`\034`".
`$!` If used in a numeric context, yields the current value of **errno**. If used in a string context, yields the corresponding error string.
`$@` The Perl error message from the last **eval** or **do** EXPR command.
`$:` The set of characters after which a string may be broken to fill continuation fields (starting with `^`) in a format.
`$0` The name of the file containing the Perl script being executed. May be assigned to.
`$$` The process ID of the currently executing Perl program. Altered (in the child process) by **fork**.
`$<` The real user ID of this process.
`$>` The effective user ID of this process.
`$(` The real group ID of this process.
`$)` The effective group ID of this process.
`$^A` The accumulator for **formline** and **write** operations.
`$^D` The debug flags as passed to perl using `-D`.
`$^F` The highest system file descriptor, ordinarily 2.
`$^I` In-place edit extension as passed to Perl using `-i`.
`$^L` Formfeed character used in formats.
`$^P` Internal debugging flag.
`$^T` The time (as delivered by **time**) when the program started. This value is used by the file test operators `-M`, `-A` and `-C`.
`$^W` The value of the `-w` option as passed to Perl.
`$^X` The name by which the currently executing program was invoked.

The following variables are context dependent and need not be localized:

`$%` The current page number of the currently selected output channel.
`$=` The page length of the current output channel. Default is 60 lines.
`$-` The number of lines remaining on the page.
`$~` The name of the current report format.
`$^` The name of the current top-of-page format.
`$|` If set to nonzero, forces a flush after every write or print on the currently selected output channel. Default is 0.
`$ARGV` The name of the current file when reading from `<>`.

The following variables are always local to the current block:

`$&` The string matched by the last successful pattern match.
`` $‘ `` The string preceding what was matched by the last successful match.
`$’` The string following what was matched by the last successful match.
`$+` The last bracket matched by the last search pattern.
`$1`...`$9`... Contain the subpatterns from the corresponding sets of parentheses in the last pattern successfully matched. `$10`... and up are only available if the match contained that many subpatterns.

Special Arrays

`@ARGV` Contains the command-line arguments for the script (not including the command name).
`@EXPORT` Names the methods a package exports by default.
`@EXPORT_OK` Names the methods a package can export upon explicit request.
`@INC` Contains the list of places to look for Perl scripts to be evaluated by the **do** FILENAME and **require** commands.
`@ISA` List of base classes of a package.
`@_` Parameter array for subroutines. Also used by **split** if not in array context.
`%ENV` Contains the current environment.
`%INC` List of files that have been included with **require** or **do**.
`%OVERLOAD` Can be used to overload operators in a package.
`%SIG` Used to set signal handlers for various signals.

HMM formula collection

Standard HMMs

Hidden Markov Process: $X_1^T = X_1, \dots, X_T$
Observed process: $Y_1^T = Y_1, \dots, Y_T$

State space: $S = \{s_1, \dots, s_N\}$
Initial probabilities: $\pi_i = P(X_1 = i),\ \ i = 1, \dots, N$
Transition probabilities: $a_{ij} = P(X_{t+1} = j | X_t = i), i, j \in S$
Emission probabilities: $b_j(Y_t \big| Y_1^{t-1}) = P(Y_t | X_t = j, Y_1^{t-1})$

$P(X_1^T, Y_1^T) = \pi_{X_1} b_{X_1}(Y_1) \prod_{t=2}^{T} a_{X_{t-1}, X_t} b_{X_t}(Y_t | Y_1^{t-1})$

Forward Algorithm

$\alpha_i(t) = P(Y_1^t, X_t = i)$

Initialization: $\alpha_i(0) = \pi_i, i = 1, \dots, N$

Tabular computation: $\alpha_i(t) = \sum_{j \in S} \alpha_j(t-1) a_{ji} b_i(Y_t | Y_1^{t-1})\, , i = 1, \dots, N, t = 1, \dots, T$

Termination: $\alpha_i(T+1) = \sum_{j \in S} \alpha_j(T) a_{ji}\, , i = 1, \dots, N$

Backward Algorithm

$\beta_i(t) = P(Y_{t+1}^T | Y_1^t, X_t = i)$

Initialization: $\beta_i(T+1) = 1, i = 1, \dots, N$

Tabular computation: $\beta_i(t) = \sum_{j \in S} \beta_j(t+1) a_{ij} b_j(Y_{t+1} | Y_1^t)\, , i = 1, \dots, N, t = T, T-1, \dots, 0$

Termination: none

The Viterbi Algorithm

$\delta_i(t) = \max_{X_1^{t-1}} P(Y_1^t, X_1^{t-1}, X_t = i)$

Initialization: $\delta_i(0) = \pi_i, i = 1, \dots, N$

Tabular computation: $\delta_i(t) = \max_{1 \le j \le N} \delta_j(t-1) a_{ji} b_i(Y_t \big| Y_1^{t-1})\, , i = 1, \dots, N, t = 1, \dots, T$
$\psi_i(t) = \mathrm{argmax}_{1 \le j \le N}\, \delta_j(t-1) a_{ji} b_i(Y_t \big| Y_1^{t-1}), i = 1, \dots, N, t = 1, \dots, T$

Termination: $\delta_i(T+1) = \max_{1 \le j \le N} \delta_j(T) a_{ji}\, , i = 1, \dots, N$
$\psi_i(T+1) = \underset{1 \le j \le N}{\mathrm{argmax}}\, \delta_j(T) a_{ji}\, , i = 1, \dots, N$

Traceback: $X_{T+1}^* = \psi_i(T+1)$
$X_t^* = \psi_{X_{t+1}^*}(t+1), t = T, T-1, \dots, 0$

The Baum-Welch Algorithm

$\gamma_i(t) = P(X_t = i \big| Y_1^T) = \frac{\alpha_i(t)\beta_i(t)}{\sum_{j\in S}\alpha_j(t)\beta_j(t)}, i = 1, \dots, N, t = 1, \dots, T$

$\xi_{ij}(t) = P(X_t = i, X_{t+1} = j \big| Y_1^T), i = 1, \dots, N, t = 1, \dots, T$

Initialization: pick arbitrary model parameters $\{\pi_i, a_{ij}, b_j : i, j = 1, \dots, N\}$

Recursion:

1. E-phase: calculate the forward and the backward algorithms for the current parameter settings.
2. M-phase: produce new parameter estimates using the re-estimation formulas below.

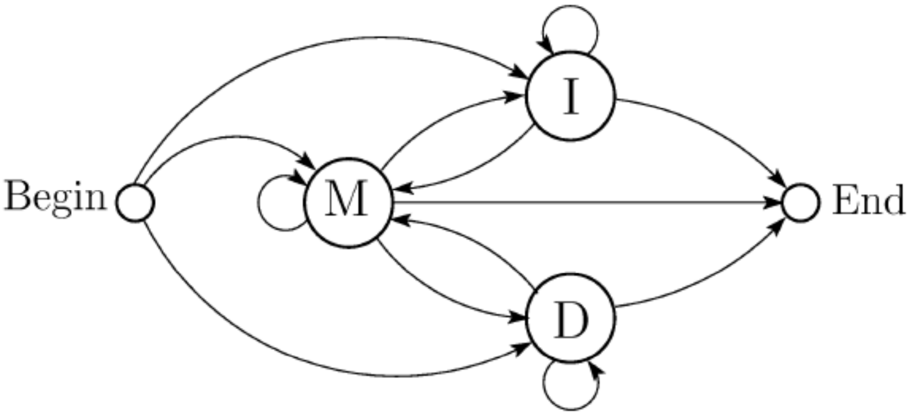
Re-estimation formulas:

$\hat{\pi}_i = \gamma_i(1)$

$$\hat{a}_{ij} = \frac{\sum_{t=1}^{T-1} \xi_{ij}(t)}{\sum_{t=1}^{T-1} \gamma_i(t)}$$

$$\hat{b}_j(c) = \frac{\sum_{t=1, Y_t = c}^{T-1} \gamma_j(t)}{\sum_{t=1}^{T-1} \gamma_j(t)}$$

Pair HMMs



State durations:

$$(d_l, e_l) = \begin{cases} (1,1) \text{ if } X_l = M \\ (1,0) \text{ if } X_l = I \\ (0,1) \text{ if } X_l = D \end{cases}$$

$$p_l = \sum_{k=1}^{L} d_k, \qquad q_l = \sum_{k=1}^{L} e_k$$

$p_0 = q_0 = 0, p_L = T, q_L = U$

The Viterbi algorithm for PHMMs

Initialization: $\delta_i(0,0) = \pi_i$

$\delta_i(t,0) = \delta_i(0,u) = 0, t > 0, u > 0$

Tabular computation:

$$\delta_M(t,u) = b_M(Y_t, Z_u \big| Y_1^{t-1}, Z_1^{u-1}) \cdot \max \begin{cases} \delta_M(t-1,u-1)a_{MM} \\ \delta_I(t-1,u)a_{IM} \\ \delta_D(t,u-1)a_{DM} \end{cases}$$

$$\delta_I(t,u) = b_I(Y_t, - \big| Y_1^{t-1}, Z_1^{u}) \cdot \max \begin{cases} \delta_M(t-1,u-1)a_{MI} \\ \delta_I(t-1,u)a_{II} \end{cases}$$

$$\delta_D(t,u) = b_D(-, Z_u \big| Y_1^{t}, Z_1^{u-1}) \cdot \max \begin{cases} \delta_M(t-1,u-1)a_{MD} \\ \delta_D(t,u-1)a_{DD} \end{cases}$$

Termination:

$$\delta_{\mathrm{M}}(T+1,U+1) = \max \begin{cases} \delta_M(T,U)a_{MM} \\ \delta_I(T,U+1)a_{IM} \\ \delta_D(T+1,U)a_{DM} \end{cases}$$

$$\delta_I(T+1,U+1) = \max \begin{cases} \delta_M(T,U)a_{MI} \\ \delta_I(T,U+1)a_{II} \end{cases}$$

$$\delta_I(T+1,U+1) = \max \begin{cases} \delta_M(T,U)a_{MD} \\ \delta_D(T+1,U)a_{DD} \end{cases}$$