

Introduction to wEMBOSS (EMBOSS)

Shahid Manzoor Adnan Niazi

SLU Global Bioinformatics Centre, Uppsala, Sweden

What is



- A free Open Source software analysis package developed for molecular biology
- Programs share a common look and feel
- Incorporates many small and large programs
- Easy to run from the command line
- Retrieval of sequence data from the web
- Easy to call from other programs(e.g. perl)
- Easy to set up behind GUIs and Web interfaces

Scope of applications

- There are many EMBOSS programs (200+)
- Many sequence analysis & display programs
- Protein 3D structure prediction being developed
- Other programs e.g enzyme kinetics

What is wEMBOSS?

 A web interface to the EMBOSS package for sequence analysis

 It's developed by Martin Sarachu (Argentina) and Marc Colet (Belgium)



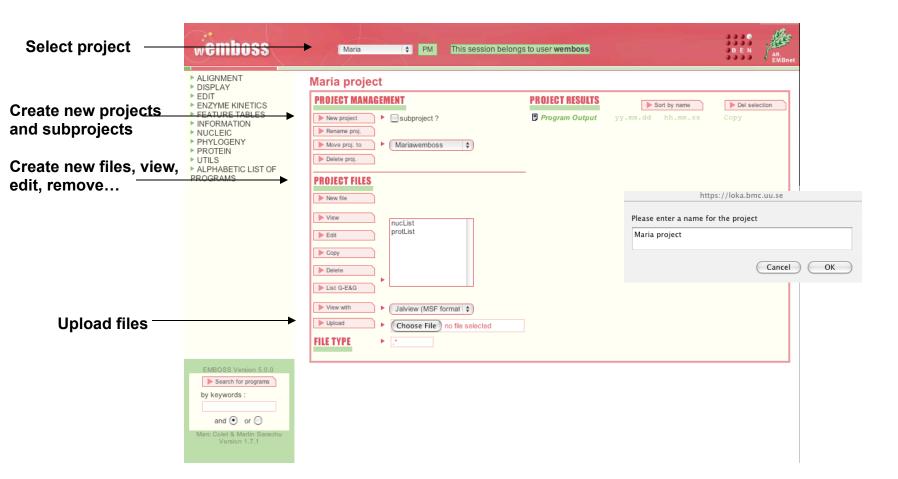
Feautures of wEMBOSS:

- Each user has a separate and private workspace.
- Organize your work by creating projects and subprojects
- Results saved for easy recover & review
- Inline help
- Keyword search for programs and documentation.



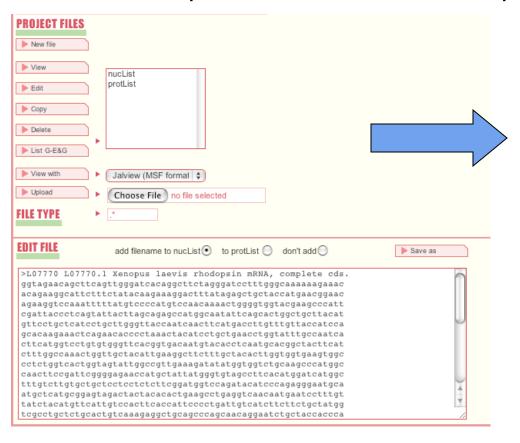
Create a project

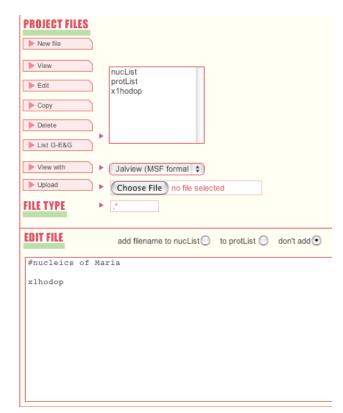
- wEMBOSS uses the concept of projects
- nucList & protList are automatically created (add names of the sequences you wish to access)



Project files

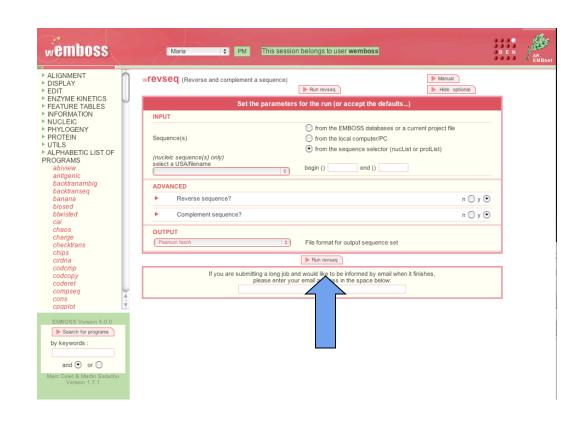
- Create a new file and paste the sequence or upload sequence
- Add sequence name to nucList or protList





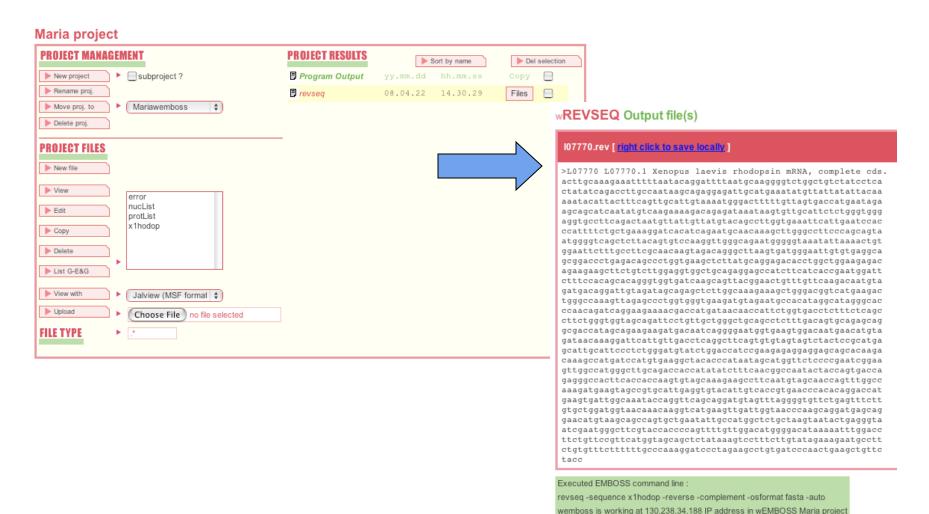
Selection of programs/files

- Drop-down menu with all available programs
- Select a program by clicking on its name.
- Choose a sequence to work with from:
 - list selector: to select a sequence from nucList or protList
 - local computer file: to upload a file from your computer
 - EMBOSS database: to access a sequence from a server

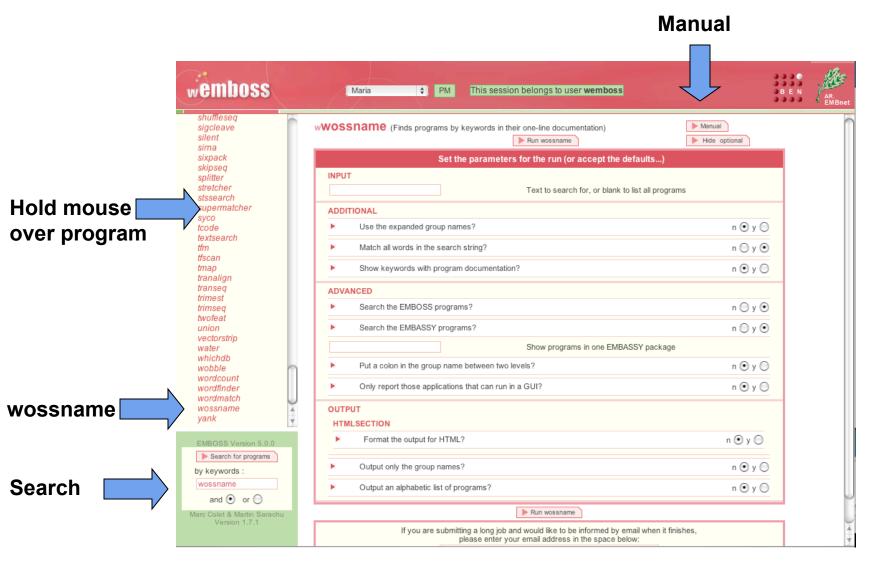


Project results

Results automatically viewed and saved

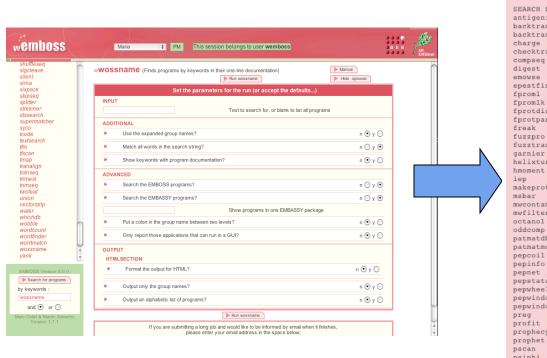


Help within wEMBOSS:



wEMBOSS program: wossname

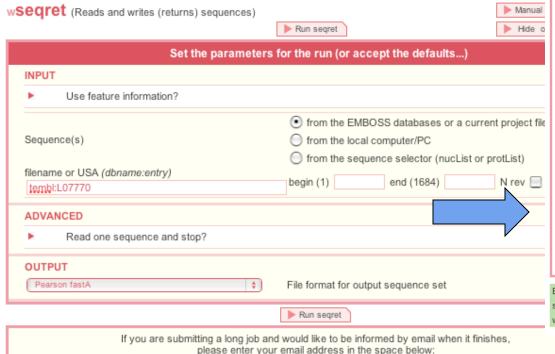
- Easy to forget a program name
- To find programs, use wossname
- wossname finds programs by looking for keywords in the description or the name of the program wossname Output file(s)



Warning SEARCH FOR 'PROTEIN' Finds antigenic sites in proteins backtranambig Back translate a protein sequence to ambiguous codons backtranseg Back translate a protein sequence Protein charge plot checktrans Reports STOP codons and ORF statistics of a protein Count composition of dimer/trimer/etc words in a sequence Protein proteolytic enzyme or reagent cleavage digest Protein identification by mass spectrometry epestfind Finds PEST motifs as potential proteolytic cleavage sites Protein phylogeny by maximum likelihood Protein phylogeny by maximum likelihood fprotdist Protein distance algorithm Protein parsimony algorithm fprotpars Residue/base frequency table or plot Protein pattern search fuzztran Protein pattern search after translation garnier Predicts protein secondary structure helixturnhelix Report nucleic acid binding motifs Hydrophobic moment calculation Calculates the isoelectric point of a protein makeprotseq Creates random protein sequences Mutate sequence beyond all recognition mwcontam Shows molwts that match across a set of files mwfilter Filter noisy molwts from mass spec output Displays protein hydropathy Find protein sequence regions with a biased composition Search a protein sequence with a motif Search a PROSITE motif database with a protein sequence patmatmotifs Predicts coiled coil regions Plots simple amino acid properties in parallel Displays proteins as a helical net pepstats Protein statistics Shows protein sequences as helices pepwheel pepwindow Displays protein hydropathy pepwindowall Displays protein hydropathy of a set of sequences Regular expression search of a protein sequence Scan a sequence or database with a matrix or profile Creates matrices/profiles from multiple alignments prophecy Gapped alignment for profiles Scans proteins using PRINTS Phi and psi torsion angles from protein coordinates psiphi shufflesed Shuffles a set of sequences maintaining composition

wEMBOSS program: seqret

- Reads in a sequence and writes it out
- Reformat sequences
- Get sequences from databases



wSEQRET Output file(s)

107770.fasta [right click to save locally]

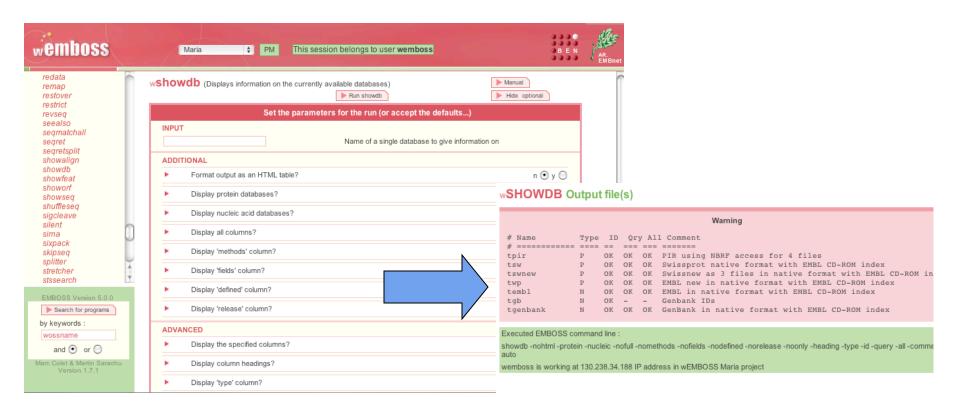
>L07770 L07770.1 Xenopus laevis rhodopsin mRNA, complete cds. ggtagaacagcttcagttgggatcacaggcttctagggatcctttgggcaaaaaagaaac acagaaggcattctttctatacaagaaaggactttatagagctgctaccatgaacggaac agaaggtccaaatttttatgtccccatgtccaacaaaactggggtggtacgaagcccatt cgattaccctcagtattacttagcagagccatggcaatattcagcactggctgcttacat gcacaagaaactcagaacacccctaaactacatcctgctgaacctggtatttgccaatca cttcatggtcctgtgtgggttcacggtgacaatgtacacctcaatgcacggctacttcat ctttggccaaactggttgctacattgaaggcttctttgctacacttggtggtgaagtggc cctctggtcactggtagtattggccgttgaaagatatatggtggtctgcaagcccatggc caacttccgattcggggagaaccatgctattatgggtgtagccttcacatggatcatggc tttgtcttgtgctgctcctctctctcggatggtccagatacatcccagagggaatgca atgctcatgcggagtagactactacacactgaagcctgaggtcaacaatgaatcctttgt tatctacatgttcattgtccacttcaccattcccctgattgtcatcttcttctgctatgg tcgcctgctctgcactgtcaaagaggctgcagcccagcaacaggaatctgctaccaccca gaaggctgagaaagaggtcaccagaatggttgttatcatggtcgttttcttcctgatctg cccagtcttcatgaccgtcccagctttctttgccaagagctctgctatctacaatcctgt catctacattgtcttgaacaacagttccgtaactgcttgatcaccaccctgtgctgtgg aaagaatccattcggtgatgaagatggctcctctgcagccacctccaagacagaagcttc ttctgtctcttccagccaggtgtctcctgcataagagcttcaccagggctgtctcagggt ccqctqcctcacacaattcccatcacttaaqccctqtctacttqttqcqaaqqcaaaqaa ttccacagttttaatatttacccccattctgcccaaccttggacactgtaagagctgacc ccattactgctgggaaggcccaagctttgttgcattctgatgtgatcctttcagcagaaa atgggtggattcaatgaatttcaccaaggctgtacataacaataacattagtctgaaggc acctcccacccagagaatgcaacacttatttatctctgtcttttcttgacatattgatgc tgcttctattcatggtcactaacaaaagtcccattttacaatgcaactgaaagtaatgt atttttgtaatataataacatatttcatgcaatctcctctgcttattggcaaggtctgat atagtgaggatagacagccagacccttgcattaaaatcctgtattaaaaatttctttgc aagt

Executed EMBOSS command line:

segret -nofeature -sequence tembl:L07770 -nofirstonly -osformat fasta -auto wemboss is working at 130.238.34.188 IP address in wEMBOSS Maria project

wEMBOSS program: showdb

Displays information of the currently available databases



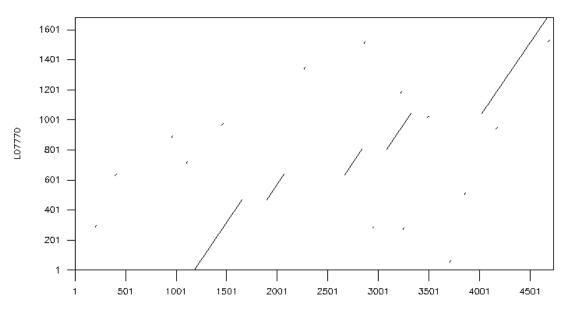
Examples of other wEMBOSS programs

- Pairwise alignment -Dotup
- Global and local alignment needle, water
- Protein translation plotorf, getorf, transeq
- Protein fingerprint pscan
- Primers eprimer3
- Microsatellites -equicktandem

Pairvise alignment - Dotup

 One sequence is represented on each axis and significant matching regions are distributed along diagonals in the matrix.

Dottup: fasta::xI23808:XL23808[1:4734] vs fasta::xIrhodo...



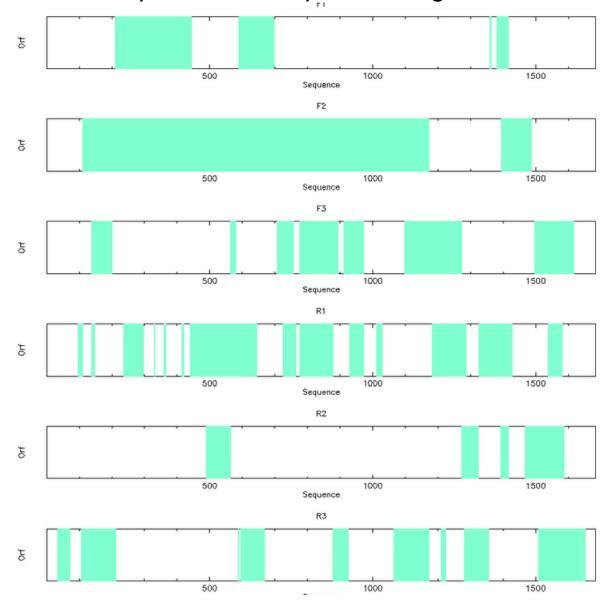
wEMBOSS program: water

Does a local alignment of two sequences

L07770	88	aggactttatagagctgctaccatgaacggaacagaaggtccaaattttt	137
BMU59922	10	AGGACTTTAAAGAGCCGCCAATATGAACGGAACAGAAGGCCCAAACTTTT	59
L07770	138	atgtccccatgtccaacaaactggggtggtacgaagcccattcgattac	187
BMU59922	60	ACATACCCATGTCCAACAAGACTGGGGTGGTACGAAGCCCATTCGAATAC	109
L07770	188	cctcagtattacttagcagagccatggcaatattcagcactg-gctgctt	236
BMU59922	110	CCTCAGTATTACCTGGCAGAGCCATGGCAATACTCCGTTTTGTGC-GCAT	158
L07770	237	acatgttcctgctcatcctgcttgggttaccaatcaacttcatgaccttg	286
BMU59922	159	ACATGTTCCTGCTCATTCTTCTTGGGTTCCCAATCAACTTCATGACCTTG	208
L07770	287	tttgttaccatccagcacaagaaactcagaacacccctaaactacatcct	336
BMU59922	209	TACGTCACCATCCAGCACAAGAAGCTCCGGACACCCTTAAACTATATCCT	258
L07770	337	gctgaacctggtatttgccaatcacttcatggtcctgtgtgggttcacgg	386
BMU59922	259	GCTGAATTTGGCCTTTGCCAACCACTTCATGGTCCTGTGTGGATTCACCG	308
L07770	387	tgacaatgtacacctcaatgcacggctacttcatctttggccaaactggt	436
BMU59922	309	TCACAATGTACTCCTCAATGAACGGATACTTCGTCTTCGGACAAACCGGT	358
L07770	437	tgctacattgaaggcttctttgctacacttggtggtgaagtggccctctg	486
BMU59922	359	TGCTATGTTGAAGGCTTCTTCGCTACCCTTGGTGGTGAAATCGCCCTTTG	408

wEMBOSS program: plotorf

• Translates sequences to 6 open reading frames.



Pick primers -eprimer3

dbxflat dbxgcg degapseg	weprimer3 (Picks PCR primers and hybridization	oligos) ▶ Run eprimer3 ▶ Hide							
descseq diffseq	Set the parameters for the run (or accept the defaults)								
digest	INPUT								
distmat dotmatcher dotpath dottup dreg edialign	The sequence from which to choose primers. The sequence must be presented 5 to 3' (nucleic sequence(s) only)	from the EMBOSS databases or a current project file from the local computer/PC from the sequence selector (nucList or protList)	3						
einverted embossdata	select a USA/filename	begin (1) end (1684) N rev							
embossuata embossversion emma	► Pick PCR primer(s)?		n ⊝ y ⊙						
emowse entret eprestfind eprimer3 equicktandem est2genome etandem	Pick PCR primers \$	Select task							
	Pick hybridization probe?		n						
	Choose File no file selected	Primer3 mispriming library file (optional) from project(s) data from local data							
extractalign extractfeat	Choose File ho he selected	II offi local data							
extractseq fclique fconsense fcontml fcontrast	ADDITIONAL PROGRAMSECTION 5 min: 0	Number of results to return							
fdiscboot fdnacomp	SEQOPTSECTION	Included region(s)							
EMBOSS Version 5.0.0		Target region(s)							
Search for programs		Excluded region(s)							
y keywords : primer		Forward input primer sequence to check							
and or		Reverse input primer sequence to check							

wEPRIMER3 Output file(s)

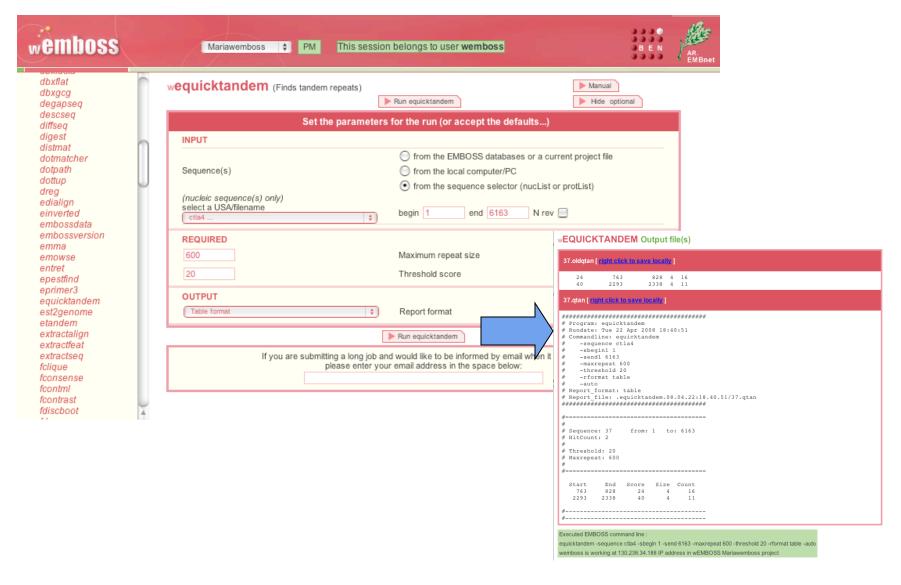
EPI	RIMER3 RE	ESULTS FOR	L07770				
			Start	Len	Tm	GC%	Sequence
1		SIZE: 199 PRIMER		20	60.00	50.00	GGCCAAACTGGTTGCTACAT
	REVERSE	PRIMER	604	20	60.14	60.00	GAGGAGGAGCACAAGAC
2		SIZE: 200 PRIMER		20	60.00	50.00	GGCCAAACTGGTTGCTACAT
	REVERSE	PRIMER	605	20	60.28	55.00	AGAGGAGGAGCACAAGA
3	FORWARD		425				GGCCAAACTGGTTGCTACAT GAGAGGAGGAGCAGCACAAG
4	FORWARD						GGCCAAACTGGTTGCTACAT AGAGAGGAGGAGCACACA
5		SIZE: 203 PRIMER	425	20	60.00	50.00	GGCCAAACTGGTTGCTACAT

Executed EMBOSS command line :

eprimer3 -sequence xIrhodop -primer -task 1 -nohybridprobe -numreturn 5 -gcclamp 0 -osize 20 -minsize 18 -maxsize 27 -otm 60.0 -mintm 57.0 -maxtm 63.0 -maxdifftm 100.0 -ogcpercent 50.0 -mingc 20.0 -maxge 80.0 -saltconc 50.0 -dnaconc 50.0 - maxpolyx 5 -productosize 200 -productsizerange 100-300 -productomintm -1000000.0 -productmintm -1000000.0 -noexplainflag -nofileflag -firstbaseindex 1 -nopickanyway -maxmispriming 12.00 -pairmaxmispriming 24.00 -numnsaccepted 0 - selfany 8.00 -selfend 3.00 -maxendstability 9.0 -auto

wemboss is working at 130.238.34.188 IP address in wEMBOSS Mariawemboss project

Find microsatellites - equicktandem



Start working on the tutorial

