

- 1) Discuss the following: Why does using protein sequences result in more accurate sequence alignments? Think about biological reasons, statistical reasons, and computational reasons.
- 2) Download the sequences you will use in the exercises (the **msa.txt** file).
  - a) Can you figure out the origins of these sequences from the identification line?
  - b) Use **T-Coffee** and **Clustal-O** to perform and compare the MSA for the given sequences (<http://www.ebi.ac.uk/Tools/msa/>). Compare the results.
    - What are the main differences? (Show some examples with screenshots)
    - Why do you think these differences occur? (No need to explain the algorithmic differences, just try to explain the main difference between the two tools).
- 3) Answer the following questions about Clustal-O.
  - a) What type of MSA method does Clustal-O use?
  - b) What is the substitution matrix used in Clustal-O?
  - c) What are the default gap opening and extension penalties for Clustal-O?
  - d) What do the ( \* ), ( : ) and ( . ) symbols at the alignment positions indicate? What does it mean if there are no symbols?

- 4) Submit the same set of sequences (upload or paste) in the **msa.txt** file to [MEME Suite](#) with default parameters.
- a) How many motifs are these sequences sharing? Is that the maximum number of motifs you can find from these alignments?
  - b) Observe the **1) seq logo**, **2) the alignment** and **3) the graphical view** of the motifs. What is the specific purpose of each representation?

Protein sequence alignments are more accurate than DNA seq. alignments, both theoretically and practically. By the means of theory, proteins are made up 20 amino acids whereas DNA letters include only 4 bases, therefore signal-to-noise ratio is much better in proteins than DNA (or RNA) sequences. In addition, due to the possibility of too many DNA mutations, there is a significant workload / computational process timing difference between protein and DNA sequences when statistically compared. Also, proteins can be observed empirically, and matrices that are empirical substitution matrices like BLOSUM62 can be used on proteins. Also, since mutations are high in DNA, phylogenetic patterns are more conserved in proteins, since proteins directly represent the phenotype of the organisms or the gene. Therefore, the mutations in the gene affect the organism's survival and fitness less than the mutations in the amino acid sequence. Amino Acid sequences, any deletions / insertions / substitutions, etc. directly affect the protein shape, therefore affect the protein function. And changes in the protein function directly changes the survival or fitness states of the organism. Since protein sequences are more conserved comparing to the DNA sequences, and DNA has higher evolving rates than proteins, converting DNA sequence to the protein sequence and then making alignment is more common technique. Also, protein sequence alignments are more beneficial in terms of the seeing amino acid replacements which is affecting the physical and chemical properties of the proteins. Substitution matrices contains empirically derived scores from amino acid sequences, and aligning non-identical amino acids are more accurate in this techniques.

sequences indicate from which organism the sequences were originally obtained

>Curvularia (P49053) Vanadium chloroperoxidase

MGSVTPIPLPKIDEPEEYNTNYILFWNHVGLLELRVTHTVGGPLTGPPLSARALGMLHLAIHDAYFSICP  
PTDFTTFLSPDTENAAAYRLPSPNGANDARQAVAGAALKMLSSLYMKPVEQPNPNPGANISDNAYAQLGLV  
LDRSVLEAPGGVDRESASFMFGEDVADVFFALLNDPRGASQEGYHPTPGRYKFDDEPTHPVVLIPVDPNN  
PNGPKMPFRQYHAPFYGKTTKRFATQSEHFLADPPGLRSNADETAEYDDAVRVAIAMGGAQALNSTKRSP  
WQTAQGLYWAYDGSNLIGTPPRFYNQIVRRIAVTYKKEEDLANSEVNNADFARLFALVDVACTDAGIFSW  
KEKWEFEFWRPLSGVRDDGRPDHGDPPFWLT LGAPATNTNDIPFKPPFPAYPSGHATFGGAVFQMVRYYN  
GRVGTWKDDEPDNIAIDMMISEELNGVNRDLRQPYDPTAPIEDQPGIVRTRIVRHFDSEWELMFENAI  
IFLGVHWRFDAAAARDILIPTTTKDVYAVDNNGATVFQNVEDIRYTTTRGTREDPEGLFPIGGVPLGIEIA  
DEIFNNGLKPTPPEIQMPQETPVQKPVGQQPVKGMWEEEQAPVVKEAP

>Embellisia (emb|CAA72344.1) vanadium chloroperoxidase

MTIDFTPVELPVVEEDAEYNWNYILFWNNVGLLELRVTHTFGALKAGPPLSPRALGMLQLAVHDAYFAIH  
PSAGFTTFLTPGAEDGAYRLPDPSYAKDARQAVAGAAIAMLSKLYMKPKVVRSPISHNAYAQLQHVLDI  
SVTKAPACDPASSSFIFGKAVATAVFDLLFHKEGADQSGYSPKPGPFKFNDPTHPVELIPVDANIPDG  
DKMPRRQYHAPYYGETAKRFGTQTEHMLADPPGIRCAGEVAEYDDAIREVYAMGGAPGLNTTKRTPHQTV  
QGMFWAYDGPKLIGTPPRLYNQIVRKIAVTYKKDNDLVNSEVNNADFARLLALVNVAMTDAGIFAWKEKW  
EFEFWRPLSGVRDDVLRDPEGKASTAAIHSGLASAPQLQNSDEAPFKPPFPAYPSGHATFGAAAFQMRK  
YYNGRLGKWATTSRDTIAVEMFVSEELNGVSRDLNPNYDPKRPITDQPGIVPTRMPRRFSSCWEMMFENA  
VSRIFLGVHWRFDAAAGQDILIPTTTKDVYAVDDKGAALFKNVEDIRYKTKGTRKGHKGLLPPIGGVPLGI  
EIANEIYNNKLSPTPPGEQMPQPPQHQQPPRRKKGELAEAKDEEQAPMMDVAP

>Drechslera (emb|CAA72008.1) vanadium chloroperoxidase

EFWRPLSGVRDDGRPDHGDPPFWLT LGAPATNTNDIPFKPPFPAYPSGHATFGGAVFQMVRYYNGRVGTW  
KDEPDNTATDMTGEELNGVNRDLRQPYDPTAPIEDQPGIVRTRIVRHFDSEWELMFENAI  
IFLGVHWRFDAAAARDILIPTTTKDVYAVDNNGATVFQNVEDIRYTTTRGTREDPEGLFPIGGVPLGIEIA  
DEIFNNGLKPTPPEIQMPQETPVQKPVGQQPVKGMWEEEQAPVVKEAP



Clustal-O is for medium or large alignments. Clustal-O uses guide tress and Hidden Markov Models to generate alignments. The default transition matrix is Gonnet, gap opening penalty is 6 bits, gap extension is 1 bit (EBI).It uses Progressive alignment construction method to make MSA. \*=conserved residue, :=conservations between groups with strong similar properties, .=conservation btw groups with weak similar properties, symbols represent the related sequences are compared and if there is no symbol this means there is no consensus sequence

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# Clustal Omega

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## Results for job clustalo-l20211205-091907-0811-35690280-p2m

Alignments

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Guide Tree

Phylogenetic Tree

Results Viewers

Submission Details

Download Alignment File

Show Colors

CLUSTAL O(1.2.4) multiple sequence alignment

Embellisia	-----	0
Curvularia	-----	0
Drechslera	-----	0

# CLUSTAL O(1.2.4) multiple sequence alignment

Embellisia	-----	0
Curvularia	-----	0
Drechslera	-----	0
Nostoc	-----	0
Corallina	-----	0
Deinococcus	-----	0
Ascophyllum	-----	0
Fucus	MLCHAADTTRGSPMPDTGVLRLTSEQRAKGWRRQLEGEKSLGFHPSETPYIKYLEGSET	60

we can see the number of hits in Clustal but not in t-coffe.

Embellisia	-----	0
Curvularia	-----	0
Drechslera	-----	0
Nostoc	-----	0
Corallina	---MGIPAD-----	6
Deinococcus	-----	0
Ascophyllum	-----	0
Fucus	WKKVKLPTDGISASKILGKIMARVRIATALAVVLAAPCLAFDEV TASGVFPEEHKHTGEG	120

Embellisia	-----	0
Curvularia	-----	0
Drechslera	-----	0
Nostoc	-----	0
Corallina	-----NLQSRKASFDTRVSAELAL-ARGVVPSLANGEELLYRNPDP	49
Deinococcus	-----	0
Ascophyllum	---QTCSTSDADDPTPPNERDDEAFASRVAAAKRELEGTGTV-----CQIN	44
Fucus	RHLQTCTNSDDALDPTAPNRRDNVAFASRRDAARRERDGTGTV-----CQIT	167

Embellisia	-----MTIDFTPVELPVVEEDAEYNWNYILFWNNVGL-----LNRVT-HTFGALKAGPP	49
Curvularia	-----MGSVTPIPLPKIDEPEEYNTNYILFWNHVGL-----LNRVT-HTVGGPLTGPP	48
Drechslera	-----	0
Nostoc	-----MPDQVINWN-----NVYLQ-TIRSNGGAP	23
Corallina	NGDPSFIVSFT-KGLPHDDNGAIIDPDDFLAFVRAINSGDEKEIADLTLPARDPDTGLP	108
Deinococcus	-----	0
Ascophyllum	NGETDLAAKFH-KSLPHDDLQV-DADAFAALEDICILNGDLSICEDVPVGNSEGD-----	97
Fucus	NGETDLATMFH-KSLPHDELQV-TADDFAILEDICILNGDFSICEDVPA----GD-----	216
Embellisia	L-SPRALGMLQLAVHDAYFAIHPSAGFTTFLTPGAEDGAYRLPDPSYAKDARQAVAGAAI	108
Curvularia	L-SARALGMLHLAIHDAYFSICPPTDFTTFLSPDTENAAYRLPSPNGANDARQAVAGAAI	107
Drechslera	-----	0
Nostoc	TWISRTGAILHSAIYDA--VNS-----IEKKYNPYLEIIPANPGASP-----EAA	66
Corallina	IWRSDLANSLELEVRGW--ENSSAGLTFDLEGPDAQSIAMPPAPVLTSP-----ELI	158
Deinococcus	-----	0
Ascophyllum	-----PVGRL---VNPTAAFAIDISGPAFSATTIPPVPTLPSP-----ELA	135
Fucus	-----PAGRL---VNPTAAFAIDISGPAFSATTIPPVPTLSSP-----ELA	254
Embellisia	AMLSKLYMKPK--VV----PRSPISHNAYAQLQH--VLDISVTKAP-----A	147
Curvularia	KMLSSLYMKPV--EQPNPNPGANISDNAYAQLGL--VLDRSVLEAP-----G	150
Drechslera	-----	0
Nostoc	AIYAAIT--VLTSDIVYPNANFPKSK-----AKNNSFFET-----ERDK	103
Corallina	AEIAELYLMALGREIEFSEFDSPKNA-EYIQFAIDQLNGLEWFNTPAMLGDPPEIRRRR	217
Deinococcus	-----	0
Ascophyllum	AQLAEVYWMALARDVPFMQYGTDDIT-VTAA---ANLAGMEGFPNLD----AVSIGSDG	186
Fucus	AQLAEVYWMALARDVPFMQYGTDEIT-TTAA---ANLAGMGGFNLD----AVSIGSDG	305

Embellisia	AC---DPASS---SFIFGKAVATAVFDLLFHKEGADQSGYSP-----KPGPFKFND	193
Curvularia	GV---DRESA---SFMFGEDVADVFFALLNDPRGASQEGYHP-----TPGRYKFDDE	196
Drechslera	-----	0
Nostoc	AIEELVSSGSVQSIGDGKELGIAAAQAIL--QNRQADGYND--NTPYTPGNQPGDWRP-	158
Corallina	GE---VTVGNLFRGILPGSEVGPYLSQYII--VGSKQIGSATGGNKTLPSPNADEFDGE	272
Deinococcus	-----	0
Ascophyllum	TV---DPLSQLFRATFVGVEVGPFISQLLV--NSF-----	216
Fucus	TV---DPFSQLFRATFVGVEVGPFVSQLLV--NSF-----	335
Embellisia	PTHPVELIPVDA---NIPDGDKMPRRQYHAPYYGETAKRFGTQTEHMLADPPGIRCA-G	248
Curvularia	PTHPVVLIPVDP---NNPNGPKMPFRQYHAPFYGKTTKRATQSEHFLADPPGLRSNAD	252
Drechslera	-----	0
Nostoc	-T--GSSAP-----VTPNWGKVK-----TFSK----API--KQFRPTRPAGFKTKKA	196
Corallina	IA--YGSITISQVRRIATPGRDFMT-----DLKV-----FL--DVQDAADFRGFESYEP	317
Deinococcus	-----	0
Ascophyllum	-T--IDSITVEPKQETFAPDVNYMV-----DFDE----WL--NIQNGGPPAGPELLDD	260
Fucus	-T--IDAITVEPKQETFAPDLNYMV-----DFDE----WL--NIQNGGPPAGPEELDE	379
Embellisia	E--VAEYDDAIREVYAMGGAPGLNTTKRTPHQTVQGMFWAYDGPKLIGTPP--RLYNQIV	304
Curvularia	E--TAEYDDAVRVAIAMGGAQALNSTKRSPWQTAQGLYNAYDGSNLIGTPP--RFYNQIV	308
Drechslera	-----	0
Nostoc	LLASLEYAAQVNEVKRLG-ADNS--TERTQEQTIDIALFWANDLDGTYKPPGHLFSITQIV	253
Corallina	G---ARLIRTIRDLATWVHFDAL--YEAYLNAC--LILLA--N-RVPFDPNIPFQQEDKL	367
Deinococcus	-----MTP--L-----GLWIEEA	11
Ascophyllum	E---LRFVRNARDLARVTFTDNI--NTEAYRGA--LILLG--L-DAFNRAVNGPFIIDID	310
Fucus	E---LRFIRNARDLARVSFVDNI--NTEAYRGS--LILLE--L-GAFSRPGINGPFIIDSD	429
Embellisia	RKIAVYKKDNDLVNSEVNNADFARLLALVNVAMTDAGIFAWKEKWEFE-----	353
Curvularia	RRIAVYKKEEDLANSEVNNADFARLFALVDVACTDAGIFSWKEKWEFE-----	357
Drechslera	-----E-----	1
Nostoc	S-----KLKGLSFYENARLFALVGLGLGDAGILAWDAKYNTD-----	290
Corallina	D-----NQDVVFNFGDAHVLSLVEVATRALKAVRYQKFNIHRRLRPEATGGL	415
Deinococcus	L-----RLGEQARLGGSDLAQVLAAT-AVAGHDAFISCWQ-----	45
Ascophyllum	R-----QAGFVNFGISHYFRLIGAA-ELAQRSSWYQKWQVHRFARPEALGGT	356
Fucus	R-----QAGFVNFGTSHYFRLIGAA-ELAQRASCYQKWQVHRFARPEALGGT	475



Embellisia	-----FWRPLSGVRDDVLRDPEGKASTAAIHSG--LASAPQLQN	390
Curvularia	-----FWRPLSGVRDDGRPDHGDPF-----W--LTLGAPATN	387
Drechslera	-----FWRPLSGVRDDGRPDHGDPF-----W--LTLGAPATN	31
Nostoc	-----LDLWRPESAIQL-----AHTDGNPGTVADPTWRPLSPN	323
Corallina	ISVNKIAAEKGESVFPEVDLAVEELEDILEKAEISNRKQNIADGDPDPD---PSFLL-P	470
Deinococcus	-----GKFEYNVAR-----PQSW---MDHVQPG	65
Ascophyllum	LHLTIKGELN---ADFDLSLLENAELLKRVAAIN---AAQNPNE---VTYLL-P	401
Fucus	LHNTIAGDLD---ADFDISLLENDELLKRVAEIN---AAQNPNE---VTYLL-P	520

. .

Embellisia	SDEAPFKPPFPAYPSGHATFGAAAFQMRKYNGRLGKWATTSRDTIAVEMFVSEELNGV	450
Curvularia	TNDIPFKPPFPAYPSGHATFGGAVFQMVRRYYNGRVGTWKDDEPDNIAIDMMISEELNGV	447
Drechslera	TNDIPFKPPFPAYPSGHATFGGAVFQMVRRYYNGRVGTWKDDEPDNIAIDMMISEELNGL	91
Nostoc	PDGTRFSPPFPAYISGHATFGAIHAGILRNFFGTDNVTFTAT-----	365
Corallina	QAFAEGSPFHPSYSGHAAVVAGACVTILKAFFDSNFQIDQVF-----	512
Deinococcus	WAPSLPTPPFPSYPSGHATVSGAAAEVLAQFFPLQA-----	101
Ascophyllum	QAIQEGSPHPSYPSGHATQNGAFATVLKALIGLDRGGDCYP-----	443
Fucus	QAIQVGSPTHPSYPSGHATQNGAFATVLKALIGLDRGGECFP-----	562

. \* . \*: \* \* \* \* . . ::

Embellisia	SRDLSNPYDPKRPITDQPGIVPTRM---PRRFSSCWEMMFENAVSRIFLGVHWRFDAAA	506
Curvularia	NRDLRQPYDPTAPIEDQPGIVRTRI---VRHFDSAWELMFENASRIFLGVHWRFDAAA	503
Drechslera	NRDLRQPYDPTAPIEDQPGIVRTRI---VRHFDSAWEMMFENASRIFLGVHWRFDAAA	147
Nostoc	-----SEDPSAR--GANGIRVT---RT-FNSFSAAALENGRSRVYLGVHYQWDADA	410
Corallina	-----EVDKDED--KLVKSSFK-GTLTVAGELNKLADNIAIGRNMAGVHYFSDQFE	560
Deinococcus	-----RQLRRDARDAAFSRVVGGIHWGVDGVA	128
Ascophyllum	-----DPVYPDDD--GLKLIDFRGSCLTFEGEINKLAVNVAFRQMLGIHYRFDGIQ	493
Fucus	-----NPVFPSSD--GLELINFEGACLTYEGEINKLAVNVAFRQMLGIHYRFDGIQ	612

~ ~ ~

Embellisia	SRDLSNPYDPKRPITDQPGIVPTRM----	PRRFSSCWEMMFENAVSRIFLGVHWRFDAAA	506
Curvularia	NRDLRQPYDPTAPIEDQPGIVRTRI----	VRHFDSAWEMLFENAIISRIFLGVHWRFDAAA	503
Drechslera	NRDLRQPYDPTAPIEDQPGIVRTRI----	VRHFDSAWEMLFENAIISRIFLGVHWRFDAAA	147
Nostoc	-----SEDPSAR--GANGIRVT--RT-FNSFSA	AALENGSRVYLGVHYQWDADA	410
Corallina	-----EVDKDED--KLVKSSFK-GTLTVAGEL	NKLADNIAIGRNMAGVHYFSDQFE	560
Deinococcus	-----RQLRRDARDAAFSRVVGGIHWGVDGVA		128
Ascophyllum	-----DPVYPDDD--GLKLIDFRGSLTFEGE	INKLAVNVAFGRQMLGIHYRFDGIQ	493
Fucus	-----NPVFPSTD--GLELINFEAGCLTYEGE	INKLAVNVAFGRQMLGIHYRFDGIQ	612

. : . . \* \* : \* \*

Embellisia	GQDIL----	IPTTKKDYYAVDDKGAALFKNVEDIRYKTKGTRK	GHKGLLPIGGV-----	556
Curvularia	ARDIL----	IPTTKKDYYAVDNNGATVFQNVEDIRYTT	RGTRREDPEGLFPIGGV-----	553
Drechslera	ARDIL----	IPTTKKDYYAVDNNGATVFQNVEDVRYSTKGT	REGREGLFPIGGV-----	197
Nostoc	AYVSGTKLADFVSE-NLLTPSVRC-----			433
Corallina	SILLGEQVAIGILEEQSLTYGENF---	FFNLP--KFDGTTIQI-----		598
Deinococcus	GLDVGQRVARALLEKRP-----			145
Ascophyllum	GLLLGETITVRTLHQELMTFAEES---	TFEFR--LFTGEVIKLFQDGTFTIDGFKC	PGLV	548
Fucus	GLLLGETITVRTLHQELMTFAEEA---	TFEFR--LFTGEVIKLFQDGTFSIDGDMC	SGLV	667

.

Embellisia	PLGIEIANEIYNNKLSPTPPGEQMPQPPQH	QGP	PPRKKGELAEAKDEEQAPMMDVAP	613
Curvularia	PLGIEIADEIFNNGLKPTPPEIQMPQETP	VQKPVGQQ-PVKGMWEEEQAPVVKEAP		609
Drechslera	PLGIEIADEIFNNGLRPTPELQMPQETP	VQKP-----	VEGMWEEEQAPIVNEAP	248
Nostoc	-----			433
Corallina	-----			598
Deinococcus	-----			145
Ascophyllum	YTGVENCVS-----			557
Fucus	YTGVAQCQA-----			676

*PLEASE NOTE: Showing colors on large alignments is slow.*

# Results for job clustalo-l20211205-091907-0811-35690280-p2m

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## Input Sequences

[clustalo-l20211205-091907-0811-35690280-p2m.input](#)

## Tool Output

[clustalo-l20211205-091907-0811-35690280-p2m.output](#)

## Alignment in CLUSTAL format with base/residue numbering

[clustalo-l20211205-091907-0811-35690280-p2m.clustal\\_num](#)

## Guide Tree

[clustalo-l20211205-091907-0811-35690280-p2m.dnd](#)

## Phylogenetic Tree

[clustalo-l20211205-091907-0811-35690280-p2m.ph](#)

## Percent Identity Matrix

[clustalo-l20211205-091907-0811-35690280-p2m.pim](#)

guide trees are different!!

Tools > Multiple Sequence Alignment > Clustal Omega

## Results for job clustalo-l20211205-091907-0811-35690280-p2m

Alignments

Result Summary

Guide Tree

Phylogenetic Tree

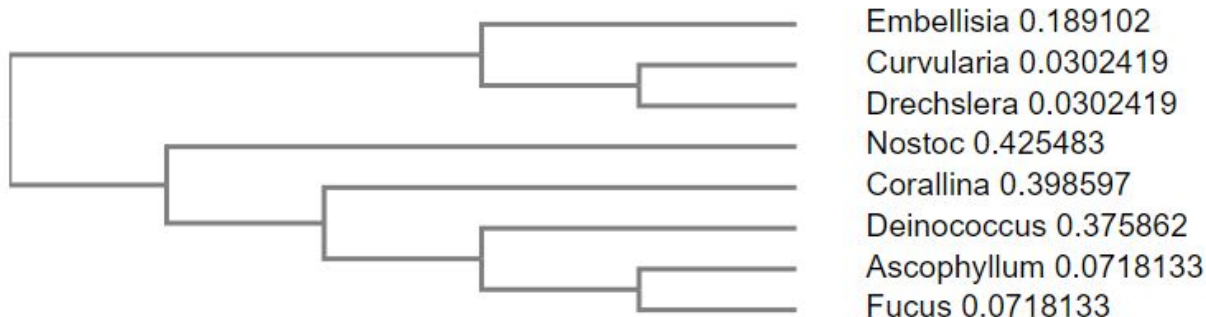
Results Viewers

Submission Details

Download Guide Tree Data

### Phylogram

Branch length: ☒ Cladogram ☐ Real





# Guide Tree

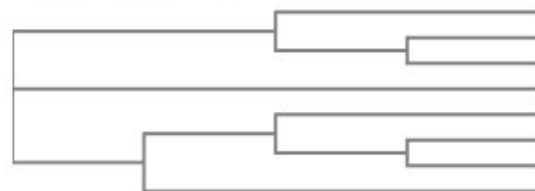
```
(  
(  
  Embellisia:0.189102  
,  
(  
  Curvularia:0.0302419  
,  
  Drechslera:0.0302419  
):0.15886  
):0.240292  
,  
(  
  Nostoc:0.425483  
,  
(  
  Corallina:0.398597  
,  
(  
  Deinococcus:0.375862  
,  
(  
  Ascophyllum:0.0718133  
,  
  Fucus:0.0718133  
):0.304049  
):0.0227354  
):0.0268853  
):0.00391147  
)  
;
```

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## Phylogenetic Tree

*This is a Neighbour-joining tree without distance corrections.*

Branch length: ☒ Cladogram ☐ Real



Embellisia 0.1744  
Curvularia 0.03365  
Drechslera 0.01473  
Nostoc 0.35951  
Corallina 0.34994  
Ascophyllum 0.06329  
Fucus 0.06871  
Deinococcus 0.34398

## Tree Data

```
(
(
(
Embellisia:0.17440,
(
(
Curvularia:0.03365,
Drechslera:0.01473)
:0.13964)
:0.20044,
Nostoc:0.35951,
(
(
Corallina:0.34994,
(
Ascophyllum:0.06329,
Fucus:0.06871)
:0.25749)
:0.06830,
Deinococcus:0.34398)
:0.04462);
```

## Results for job clustalo-I20211205-091907-0811-35690280-p2m

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## Program

clustalo

## Version

1.2.4

## Number of Sequences

8

## Launched Date

Sun, Dec 05, 2021 at 09:19:08

## End Date

Sun, Dec 05, 2021 at 09:19:10

## Input Sequences

[clustalo-I20211205-091907-0811-35690280-p2m.input](#)

## Output Result

[clustalo-I20211205-091907-0811-35690280-p2m.output](#)

## Command

```
$APPBIN/clustal-omega-1.2.4/bin/clustalo --infile clustalo-I20211205-091907-0811-35690280-p2m.sequence --threads 8 --MAC-  
RAM 8000 --verbose --guidetree-out clustalo-I20211205-091907-0811-35690280-p2m.dnd --outfmt clustal --resno --outfile  
clustalo-I20211205-091907-0811-35690280-p2m.clustal_num --output-order tree-order --seqtype protein
```

## Input Parameters

## Output guide tree

true

## Output distance matrix

false

input parameters are more higher in clustal because it based on HMM but input parameters are low in t-coffee since it based on heuristic algorithms. it affects the general output and phylogeny of the sequences. Also guide trees are different. T-coffee can be detect patterns whereas Clustal can be detect conserved areas more efficiently.

Output guide tree	true
Output distance matrix	false
Dealign input sequences	false
mBed-like clustering guide tree	true
mBed-like clustering iteration	true
Number of iterations	0
Maximum guide tree iterations	-1
Maximum HMM iterations	-1
Output alignment format	clustal_num
Output order	aligned
Sequence Type	protein



T-Coffee is used for small alignments. In T-coffee, comparison is made by creating two libraries for both global (with ClustalW) and local (with Lalign) and uses heuristic algorithms on this libraries. T-coffee motifs are good at identifying.

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# T-Coffee

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[Tools](#) > [Multiple Sequence Alignment](#) > [T-Coffee](#)

## Results for job [tcoffee-l20211205-092003-0880-78434929-p1m](#)

[Alignments](#)[Result Summary](#)[Guide Tree](#)[Phylogenetic Tree](#)[Results Viewers](#)[Submission Details](#)[Download Alignment File](#)[Show Colors](#)

CLUSTAL W (1.83) multiple sequence alignment

Ascomphyllum	QTCS-----T-----
Corallina	MGIP-----A-----
Curvularia	MGSV-TPIPLPKIDEPEEYNTNYILFWNHVGLNLRVTHTVGGPLTGPPL
Deinococcus	MTPL---GLWI-----
Drechslera	-----
Embellisia	MTIDFTPVELPVVEEDA EYNNWNYILFWNNVGLNLRVTHTFGALKAGPPL
Fucus	MLCH-----A-----
Nectoc	MDP-----QVTHNNVAYLQTTFS-----NGCAPTH

# CLUSTAL W (1.83) multiple sequence alignment

```

Ascophyllum      QTCS-----T-----
Corallina        MGIP-----A-----
Curvularia      MGSV-TPIPLPKIDEPEEYNTNYILFWNNHVGLELN RVTHTVGGPLTGPPL
Deinococcus      MTPL----GLWI-----
Drechslera       -----
Embellisia       MTIDFTPVELPVVEEDAEYNWNYILFWNNVGLELN RVTHTFGALKAGPPL
Fucus            MLCH-----A-----
Nostoc           MPD-----QVINWNNVYLQTI RS-----NGGAPT W
    
```

the order of the sequences  
(organisms) is different  
than culstal

```

Ascophyllum      -----
Corallina        -----
Curvularia      SARALGMLHLAIHDAYFSICPPTDFTTFLSPDTENAAYRLP-SPNGANDA
Deinococcus      -----
Drechslera       -----
Embellisia       SPRALGMLQLAVHDAYFAIHPSAGFTTFLTPGAEDGAYRLP-DPSYAKDA
Fucus            -----ADT-TRGSPMPDTGVLRLLTSEQRAKGW
Nostoc           ISRTGAILHSAIYDAVNSIEKKYNP--YLE-----IIP-ANPGASPE
    
```

```

Ascophyllum      -----
Corallina        -----
Curvularia      RQAVAGA-----
Deinococcus      -----
Drechslera       -----
Embellisia       RQAVAGA-----
Fucus            RRQLEGEKSLGFHPSETPYIKYLEGSETWKKVKLPTDGISASKILGKIMA
Nostoc           AAAIYAA-----
    
```

```

Ascophyllum      -----
Corallina        -----
Curvularia      -----ALKMLSSLYMKPVEQPNPNPGAN-----
Deinococcus      -----E-----
Drechslera       -----
Embellisia       -----AIAMLSKLYMKPKVVP---RSP-
Fucus            RVRIATALAVVLAAPCLAFDEVTASGVFP---EEHKHTGEGRHLQTCTN
Nostoc           -----YTVLTSDIVYPNANFP---KSK-----
    
```

Ascomycetum	SDDADDPTPPNERDDEAFASRV--AAAKRELEGTGTVCQI--NNG-----
Corallina	DN-----LQSRKASFDTRV--SAAELAL-ARGVVPSL-ANGEELLYR
Curvularia	-----ISDNAYAQLGLVLDLRSVLEA-PGGVDRES--AS-----
Deinococcus	-----
Drechslera	-----
Embellisia	-----ISHNAYAQLQHVLDISVTKA-PAACDPAS--SS-----
Fucus	SDDALDPTAPNRRDNVAFASRR--DAARRERDGTGTVCQI--TNG-----
Nostoc	-----AKNNSFFETER--DKAIEELVSSGVSQSIGDG-----

Ascomycetum	-----ETDLAAKFHKSLPHDDLQ--VDADAFALDILNGDLSICED
Corallina	NPDPEGDPSPFIVSFTKGLPHDNGAIIDPDDFLAFVRAINSGDEKEIAD
Curvularia	-----FMFGE-DVAD--VFFALLNDPRGASQEGY
Deinococcus	-----
Drechslera	-----
Embellisia	-----FIFGK-AVAT--AVFDLLFHKEGADQSGY
Fucus	-----ETDLATMFHKSLPHDELQ--VTADDFAILEDILNGDFSICED
Nostoc	-----KELGI-AAQ--AILQNRQADGYNDNTPY

Ascomycetum	--VPVGNSEGD-----VGRNVNPTAAFAIDISGPAF
Corallina	--LTLGP-ARDPDTGLPIWRSDLANSLELVRGWENSAGLTFDLEGDA
Curvularia	HPTPG-RYKFD-----DE-----PTH
Deinococcus	-----
Drechslera	-----
Embellisia	SPKPG-PFKFN-----DE-----PTH
Fucus	--VPAG--DP-----AGRLNVNPTAAFAIDISGPAF
Nostoc	--TPG-NQPGD-----WR-----PTG

Ascomycetum	SAT-TIPPVPTLPSPELAAQLAEVYWMALARDVPFMQY-----
Corallina	QSI-AMPPAPVLTSPELIAEIAELYMALGREIEFSEFSDPKNAEYIQ-F
Curvularia	PVV-LIPVDN-----NPNPKMPFRQYHAPFYGKTTKRF
Deinococcus	-----EALRL
Drechslera	-----
Embellisia	PVE-LIPVDN-----IPDGDKMPRRQYHAPYYGETAKRF
Fucus	SAT-TIPPVPTLPSPELAAQLAEVYWMALARDVPFMQY-----
Nostoc	SSAPVTPNWGK-----VKTFKAPIKQFRP-----

Ascomyllum	GTDDITVTAAANLAGMEGFPNLDAVSIGSDGTVDPLSQLFRATFV-GVET
Corallina	AIDQLNGLEWFNTPAMLGDPPEIRRRRGEVTV---GNLFRGILP-GSEV
Curvularia	A-----TQSEHFLADPPG-----LRNADET
Deinococcus	G-----
Drechslera	-----
Embellisia	G-----TQTEHMLADPPG-----IRC-AGEV
Fucus	GTDEITTTAAANLAGMGGFPNLDAVSIGSDGTVDPPFSQLFRATFV-GVET
Nostoc	-----TRPAG-----FKTKKALL

Ascomyllum	--GPFISQLLV-N-----SFTIDSITVEPK
Corallina	--GPYLSQYIIVGSKQIGSATGGNKTIVSPNAADEFDGEIAYGSITISQR
Curvularia	--AEYDD--AVRVAIAMGGAQALNSTKRSPWQTAQ-----
Deinococcus	-----EQARL-----
Drechslera	-----
Embellisia	--AEYDD--AIREVYAMGGAPGLNTTKRTPHQTVQ-----
Fucus	--GPFVSQLLV-N-----SFTIDAITVEPK
Nostoc	ASLEYAA--QVNEVKRLGA---DNSTERTQEQTDI-----

Ascomyllum	QETFAPDVNYMVDFFDEWLNIQNGGPPAGPELDDDELRFVRNARDLARVTF
Corallina	VRIATPGRDFMTDLKVFLDVQDAADFGRGFESYEPGARLIRTIRDLATWVH
Curvularia	-----GLYW---AY--DGSNLIGTPPRFYN-----
Deinococcus	-----
Drechslera	-----
Embellisia	-----GMFW---AY--DGPKLIGTPPRLYN-----
Fucus	QETFAPDLNYMVDFFDEWLNIQNGGPPAGPEELDEELRFIRNARDLARVSF
Nostoc	-----ALFW---AN--DLGTYKPPGHLFSI-----

Ascomyllum	TDNINTEAYRGA-LILLGLDAFNRAVNGPFID--IDRQAGFVNFGISH
Corallina	FDALY-EAYLNACLILLANRVFPDP--NIPFQQEDKLDNQDVFNFGDAH
Curvularia	-----QIVRRI--AVTYKKEEDLANSEV--NNADFAR
Deinococcus	-----GGSDLAQ
Drechslera	-----
Embellisia	-----QIVRKI--AVTYKKDNDLVNSEV--NNADFAR
Fucus	VDNINTEAYRGS-LILLELGAFSRPGINGPFID---SDRQAGFVNFGTSH
Nostoc	-----TQIVSKL--KG-----L-SFYENAR



Ascophyllum	YFRLIG-AAELAQRSSWYQKWQ--VHRFARPEALGGTLHL-TIKGE----
Corallina	VLSLVTEVATRALKAVRYQKFN--IHRRLRPEATGGLISVNKIAAEKGES
Curvularia	LFALVDVACTDAGIFSWKEKWE--F-EFWRPLSGV--R-----
Deinococcus	VLAATAVAGHDAFISCWQGKFE--Y-NVARPQSWM-----
Drechslera	-----EFWRPLSGV--R-----
Embellisia	LLALVNVAMTDAGIFAWKEKWE--F-EFWRPLSGV--R-----
Fucus	YFRLIG-AAELAQRASCYQKWQ--VHRFARPEALGGTLHN-TIAGD----
Nostoc	LFALVGLGLGDAGILAWDAKYNTDL-DLWRPESAI--Q-----

\*\* ;

Ascophyllum	LNADFDSLLENAELLK-----RVAAINAAQNPNN----EVTYLLPQAI
Corallina	VFPEVDLAVEELEDILEKAEISNRKQNI-ADGDPDP----DPSFLLPQAF
Curvularia	-----DDGR-----PD-HGD-----PFWLTLGAPA-TN-TND
Deinococcus	-----D-HVQ-----PGW----AP-----
Drechslera	-----DDGR-----PD-HGD-----PFWLTLGAPA-TN-TND
Embellisia	-----DDVLRDPEGKASTAAI-----HSGLAS-APQLQN-SDE
Fucus	LDADFDISLLENDELK-----RVAEINAAQNPNN----EVTYLLPQAI
Nostoc	-----LAHTDGN-----PGTVAD-----PTW----RPLSPN-PDG

Ascophyllum	QEGSPTHPSYPSGHATQNGAFATVLKALIGLD-----
Corallina	AEGSPFHPSYGSGHAVVAGACVTILKAFFDSN-----
Curvularia	IPFKPPFPAYPSGHATFGGAVFQMVRYYNGRVGTWKDDEPDNIAIDMMI
Deinococcus	SLPTPPFPSYPSGHATVSGAAAEVLAQFFPLQARQLR-----
Drechslera	IPFKPPFPAYPSGHATFGGAVFQMVRYYNGRVGTWKDDEPDNIAIDMMI
Embellisia	APFKPPFPAYPSGHATFGAAAFQMVRKYNGRLGKWATTSRDTIAVEMFV
Fucus	QVGSPTHPSYPSGHATQNGAFATVLKALIGLD-----
Nostoc	TRFSPFPAYISGHATFGAIHAGILRNFFGTDNVTFTATSE-----

\* , \* : \* \*\*\*\* . . ::

Ascophyllum	-----RGGDCYPDPVYPDDGLKLIDFRGSCLTFEGEINKLAVNNAF
Corallina	-----FQIDQVF-EVDKDEDKLVKSSFKGT-LTVAGELNKLADNIAI
Curvularia	SEELNGVNRLRQPYDPTAPIEDQPGIVRTRI-VRHFD-SAWELMFENAI
Deinococcus	-----RDARDAAF
Drechslera	SEELNGLNRDLRQPYDPTAPIEDQPGIVRTRI-VRHFD-SAWEMMFENAI
Embellisia	SEELNGVSRDLNPNYDPKRPITDQPGIVPTRM-PRRFS-SCWEMMFENAV
Fucus	-----RGGECFPNPVFPSSDGLLELINFEGACLTYEGEINKLAVNNAF
Nostoc	-----DPSAR-----GANGIRV-TRTFN-SFSAAALENGR

: .

Ascophyllum	GRQMLGIHYRFDGIQGLLLGETITVRTLHQELMTFAEESTFEFRL-----
Corallina	GRNMAGVHYFSDQFESILLGEQVAIGILEEQSLTYGENFFFNLPK-----
Curvularia	SRIFLGVHWRFDAAAARDI----LIPTTTTKDQVAVDNNNGATVFQNVEDIR
Deinococcus	SRVVGGIHWGVDGVAGLDV-----
Drechslera	SRIFLGVHWRFDAAAARDI----LIPTTTTKDQVAVDNNNGATVFQNVEDVR
Embellisia	SRIFLGVHWRFDAAAAGQDI----LIPTTKDQVAVDDKGAALFKNVEDIR
Fucus	GRQMLGIHYRFDGIQGLLLGETITVRTLHQELMTFAEEATFEFRL-----
Nostoc	SRVYLVGHYQWDADAAYVS-----

. \*    \*: \*:    \*    .

Ascophyllum	FTGEVIKLFQDGTFTIDGFKCPLVYTGVENCV-----
Corallina	FDGTTIQ-----
Curvularia	YTTRGTREDPEGLFPIGG-----VPLGIEIADEIFNNGLKPTPPEIQPM
Deinococcus	-----GQRVARALL-----
Drechslera	YSTKGTRGREGLFPIGG-----VPLGIEIADEIFNNGLRPTPELQPM
Embellisia	YKTKGTRKGHKGLLPPIGG-----VPLGIEIANEIYNNKLSPTPPGEQPM
Fucus	FTGEVIKLFQDGTFSIDGDMCSGLVYTGVAQC-----
Nostoc	-----GTKLADFVSENLLTPSVR-----

Ascophyllum	-----S
Corallina	-----I
Curvularia	PQETPVQKPVGQQ-PVKGMWEEEQAPVVKAP
Deinococcus	-----EKRP
Drechslera	PQETPVQKP-----VEGMWEEEQAPIVNEAP
Embellisia	PQPPQHQPGRKKGELAEAKDEEQAPMDVAP
Fucus	-----A
Nostoc	-----C

*PLEASE NOTE: Showing colors on large alignments is slow.*

Tools > Multiple Sequence Alignment > T-Coffee

## Results for job tcoffee-l20211205-092003-0880-78434929-p1m

[Alignments](#)[Result Summary](#)[Guide Tree](#)[Phylogenetic Tree](#)[Results Viewers](#)[Submission Details](#)

### Input Sequences

[tcoffee-l20211205-092003-0880-78434929-p1m.input](#)

### Tool Output

[tcoffee-l20211205-092003-0880-78434929-p1m.output](#)

### Alignment in CLUSTAL format

[tcoffee-l20211205-092003-0880-78434929-p1m.clustalw](#)

### Guide Tree

[tcoffee-l20211205-092003-0880-78434929-p1m.dnd](#)

### Phylogenetic Tree

[tcoffee-l20211205-092003-0880-78434929-p1m.ph](#)

### Percent Identity Matrix

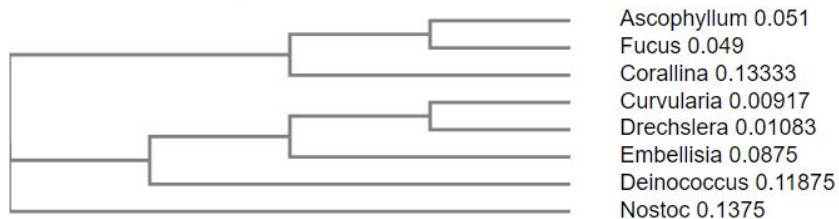
[tcoffee-l20211205-092003-0880-78434929-p1m.pim](#)

## Results for job tcoffee-l20211205-092003-0880-78434929-p1m

[Alignments](#)[Result Summary](#)[Guide Tree](#)[Phylogenetic Tree](#)[Results Viewers](#)[Submission Details](#)[Download Guide Tree Data](#)

### Phylogram

Branch length: ☒ Cladogram ☐ Real



### Guide Tree

```
((((Ascophyllum:0.05100,Fucus:0.04900):0.07167,Corallina:0.13333):0.00750,(((Curvularia:0.00917,Drechslera:0.01083):0.08250,Embellisia:0.08750):0.02875,Deinococcus:0.11875):0.00750,Nostoc:0.13750);
```



Tools > Multiple Sequence Alignment > T-Coffee

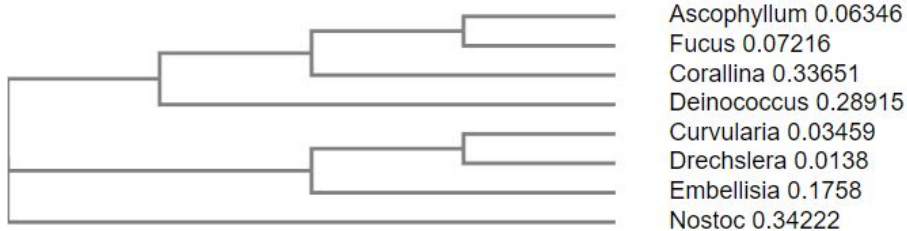
## Results for job tcoffee-I20211205-092003-0880-78434929-p1m

[Alignments](#)[Result Summary](#)[Guide Tree](#)[Phylogenetic Tree](#)[Results Viewers](#)[Submission Details](#)[Download Phylogenetic Tree Data](#)

### Phylogenetic Tree

*This is a Neighbour-joining tree without distance corrections.*

Branch length: ☒ Cladogram ☐ Real



guide trees and phylogenetic trees are different in t-coffee and clustal!!  
because the approaches of to align the sequences are different &&  
T-coffee motifs are good at identifying.

# Tree Data

```
(  
(  
(  
(  
Ascophyllum:0.06346,  
Fucus:0.07216)  
:0.23498,  
Corallina:0.33651)  
:0.10449,  
Deinococcus:0.28915)  
:0.04356,  
(  
(  
Curvularia:0.03459,  
Drechslera:0.01380)  
:0.13184,  
Embellisia:0.17580)  
:0.16521,  
Nostoc:0.34222);
```

---

## Results for job tcoffee-I20211205-092003-0880-78434929-p1m

Alignments Result Summary Guide Tree Phylogenetic Tree Results Viewers **Submission Details**

<b>Program</b>	<b>Number of Sequences</b>	<b>Launched Date</b>	<b>Input Sequences</b>
T-COFFEE	8	Sun, Dec 05, 2021 at 09:11:42	<a href="#">tcoffee-I20211205-092003-0880-78434929-p1m.input</a>
<b>Version</b>		<b>End Date</b>	<b>Output Result</b>
13.41.0.28bdc39		Sun, Dec 05, 2021 at 09:11:48	<a href="#">tcoffee-I20211205-092003-0880-78434929-p1m.output</a>

## Command

```
$APPBIN/tcoffee:13.45.0.4846264 /t_coffee -in tcoffee-I20211205-092003-0880-78434929-p1m.sequence -case=upper -n_core=8 -  
output=clustalw,msf,phylip,score_html,fasta -outorder=aligned -type=protein; echo ' '
```

## Input Parameters

Alignment format

clustalw

Matrix

none

Output Order

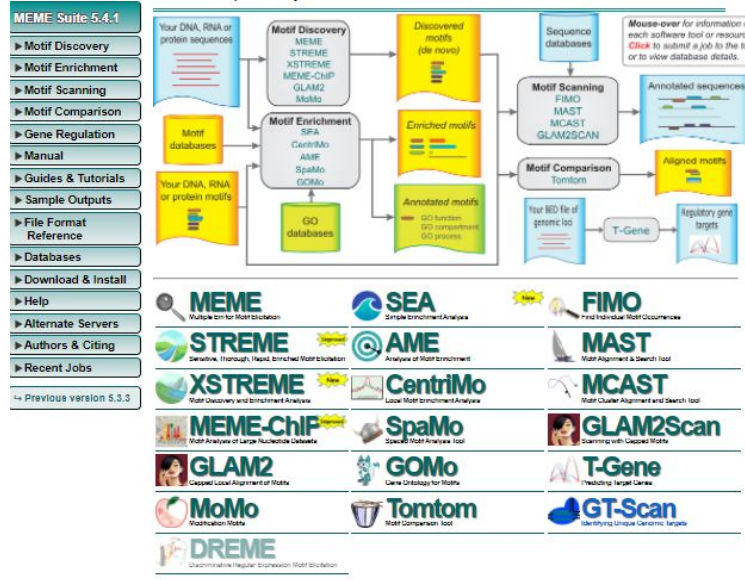
aligned

Sequence Type

protein

# The MEME Suite

Motif-based sequence analysis tools



Maintenance and development of the MEME Suite is funded by grant R01 GM103544 from the National Institutes of Health.

Developed and maintained by



University of Nevada, Reno



THE UNIVERSITY OF QUEENSLAND AUSTRALIA







# MEME

Multiple Em for Motif Elicitation

## MEME Suite 5.4.1

► Motif Discovery

► Motif Enrichment

► Motif Scanning

► Motif Comparison

► Gene Regulation

► Manual

► Guides & Tutorials

► Sample Outputs

► File Format  
Reference

► Databases

► Download & Install

► Help

► Alternate Servers

► Authors & Citing

► Recent Jobs

↩ Previous version 5.3.3

Please wait. Your MEME job is now queued awaiting available resources. You may bookmark this page or use the **Recent Jobs** menu at the left to access your job's results.

## Job Details ...

## Results

- [\(Primary\) Sequences](#)

## Status Messages

- Parsing arguments
- Arguments ok
- Starting meme  
meme msa.txt -protein -oc . -nostatus -time 14400 -mod zoops -nmotifs 3 -minw 6 -maxw 50 -objfun classic -markov\_order 0

3 motifs are found.  
seq logo: is generated based on a set of  
frequency matrices



For further information on how to interpret these results please access <https://meme-suite.org/meme/doc/meme.html>.  
To get a copy of the MEME software please access <https://meme-suite.org>.

If you use MEME in your research, please cite the following paper:

Timothy L. Bailey and Charles Elkan, "Fitting a mixture model by expectation maximization to discover motifs in biopolymers", *Proceedings of the Second International Conference on Intelligent Systems for Molecular Biology*, AAAI Press, Menlo Park, California, 1994. [\[full text\]](#)

[DISCOVERED MOTIFS](#) | [MOTIF LOCATIONS](#) | [INPUTS & SETTINGS](#) | [PROGRAM INFORMATION](#) | [RESULTS IN TEXT FORMAT](#)  | [RESULTS IN XML FORMAT](#) 

## DISCOVERED MOTIFS

	Logo 	E-value 	Sites 	Width 	More 	Submit/Download 
1.		1.8e-071	8	40		
2.		1.1e-046	7	27		
3.		2.8e-026	3	30		

# DISCOVERED MOTIFS



A motif is a sequence pattern that occurs repeatedly in a group of related sequences and here is the motif patterns of the alignments

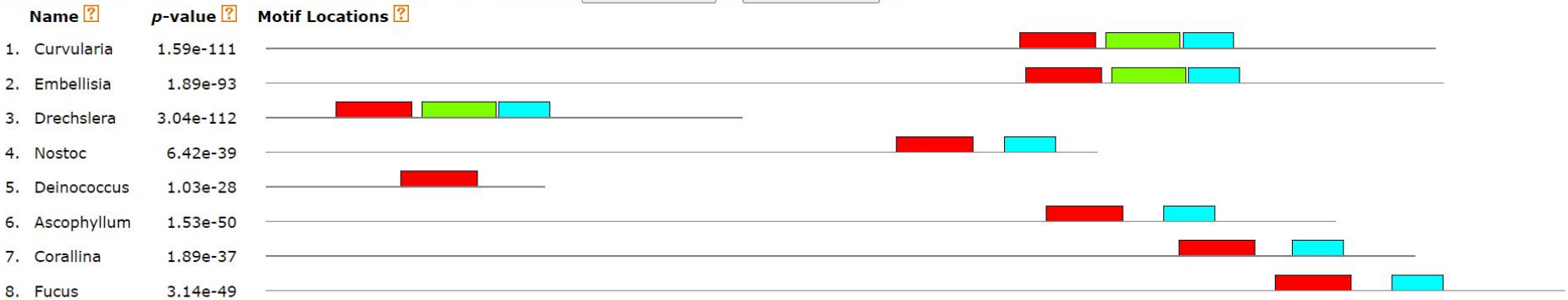




the location of the motifs that are found in the sequences with p values (significance values)




## MOTIF LOCATIONS

☒ Only Motif Sites ☐ Motif Sites+Scanned Sites ☐ All Sequences [Download PDF](#) [Download SVG](#)



# INPUTS & SETTINGS




## Sequences

Role	Source 	Alphabet 	Sequence Count 	Total Size 
Primary Sequences	msa.txt	Protein	8	3879

## Background Model

**Source:** built from the (primary) sequences

**Order:** 0

	Name 	Freq. 	Bg. 
<b>A</b>	Alanine	0.104	0.104
<b>C</b>	Cysteine	0.00722	0.00722
<b>D</b>	Aspartic acid	0.0693	0.0694
<b>E</b>	Glutamic acid	0.0611	0.0611
<b>F</b>	Phenylalanine	0.0534	0.0534
<b>G</b>	Glycine	0.0809	0.081
<b>H</b>	Histidine	0.0183	0.0183
<b>I</b>	Isoleucine	0.0526	0.0526
<b>K</b>	Lysine	0.0338	0.0338
<b>L</b>	Leucine	0.082	0.082
<b>M</b>	Methionine	0.0162	0.0162
<b>N</b>	Asparagine	0.048	0.048
<b>P</b>	Proline	0.0755	0.0755
<b>Q</b>	Glutamine	0.0351	0.0351
<b>R</b>	Arginine	0.0518	0.0518
<b>S</b>	Serine	0.0498	0.0498
<b>T</b>	Threonine	0.0596	0.0596
<b>V</b>	Valine	0.0606	0.0606
<b>W</b>	Tryptophan	0.0137	0.0137
<b>Y</b>	Tyrosine	0.0276	0.0276

max. number of motifs are 3

#### Other Settings

<b>Motif Site Distribution</b>	ZOOPS: Zero or one site per sequence
<b>Objective Function</b>	E-value of product of p-values
<b>Starting Point Function</b>	E-value of product of p-values
<b>Site Strand Handling</b>	This alphabet only has one strand
<b>Maximum Number of Motifs</b>	3
<b>Motif E-value Threshold</b>	no limit
<b>Minimum Motif Width</b>	6
<b>Maximum Motif Width</b>	50
<b>Minimum Sites per Motif</b>	2
<b>Maximum Sites per Motif</b>	8

[Show Advanced Settings](#)

#### MEME version

5.4.1 (Release date: Sat Aug 21 19:23:23 2021 -0700)

#### Reference

Timothy L. Bailey and Charles Elkan, "Fitting a mixture model by expectation maximization to discover motifs in biopolymers", *Proceedings of the Second International Conference on Intelligent Systems for Molecular Biology*, pp. 28-36, AAAI Press, Menlo Park, California, 1994. [\[full text\]](#)

#### Command line

```
meme msa.txt -protein -oc . -nostatus -time 14400 -mod zoops -nmotifs 3 -minw 6 -maxw 50 -objfun classic -markov_order 0
```

# References

Wernersson, R., & Pedersen, A. G. (2003). RevTrans: Multiple alignment of coding DNA from aligned amino acid sequences. *Nucleic acids research*, 31(13), 3537–3539. <https://doi.org/10.1093/nar/gkg609>

<https://www.ebi.ac.uk/seqdb/confluence/display/THD/Help+-+Clustal+Omega+FAQ#HelpClustalOmegaFAQ-What+is+ClustalOmega?>

<https://user.ceng.metu.edu.tr/~tcan/ceng465/Schedule/MSAComparison.pdf>