

LOGO

链球菌属噬菌体Gp3蛋白结构的 预测



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芳、郑莹莹

1 背景介绍

2 gp3氨基酸序列分析 Blast和同源建模

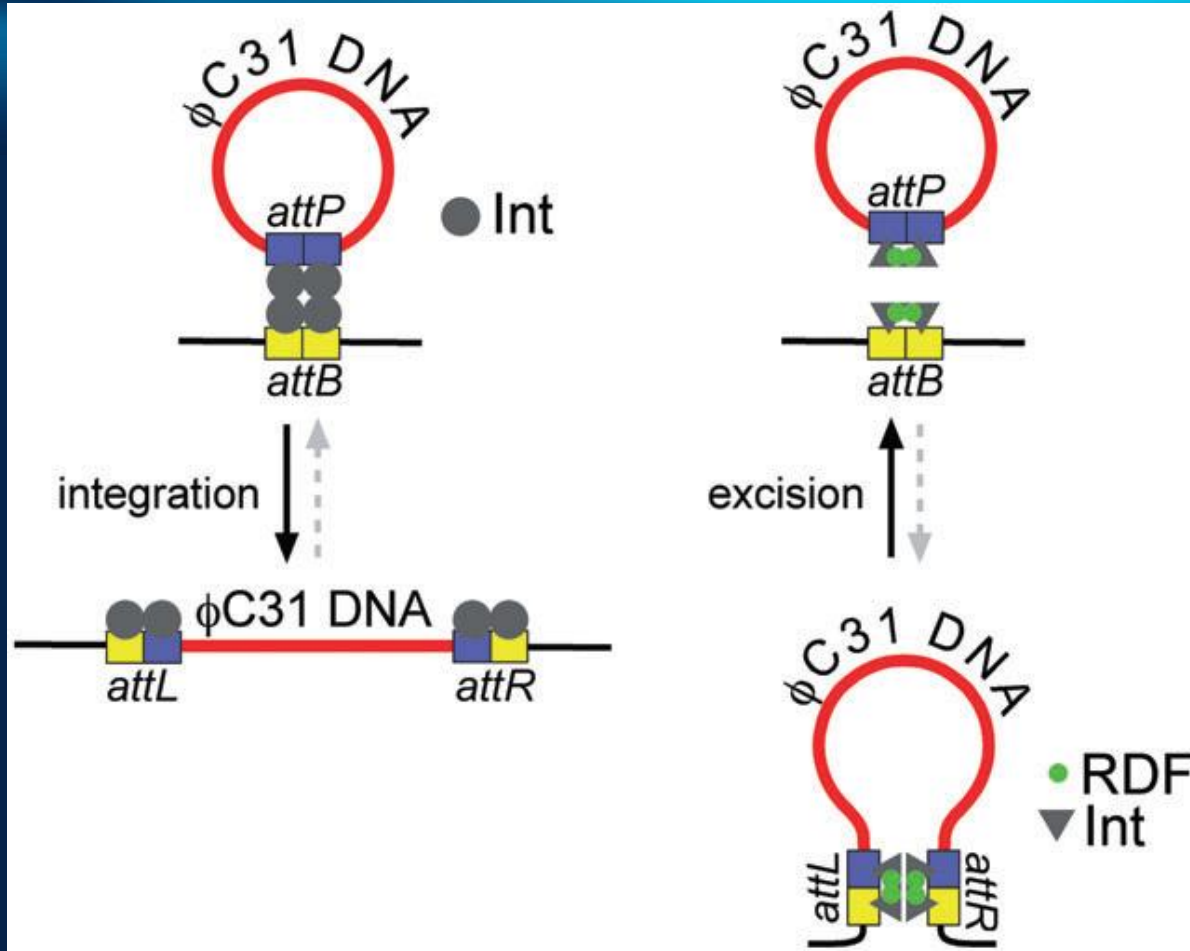
3 gp3和gp47比较

4 结论和参考文献



来自于链霉菌噬菌体 ϕ C31的丝氨酸整合蛋白Int可用于调节噬菌体基因组在寄主染色体中的插入和删除。

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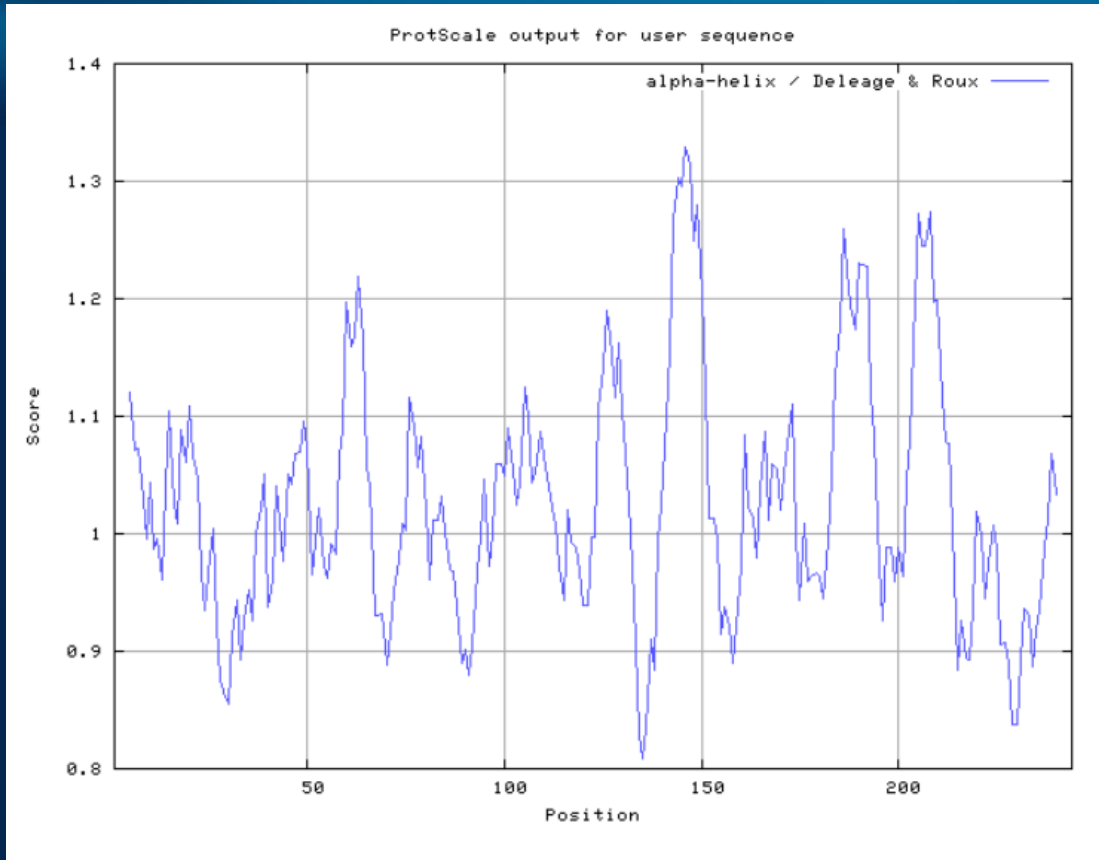


ϕ C31整合和删除机制：Int亚基中控制插入的用灰色圆圈表示，控制删除的用三角形表示，Int亚基用于预测研究蛋白的构想改变，Int和RDF的亚基仍然不清楚，黑线和红线代表了宿主和噬菌体基因组DNA。

- fC31表达早期蛋白gp3（RDF家族成员），当有gp3存在时，整合酶能催化切离反应，当没有gp3时，整合酶只能催化整合反应。
- 有文章表明整合酶的方向性由一个预测的卷曲螺旋结构控制的，我们猜想gp3中存在卷曲螺旋结构。
- fC31 gp3的功能特点类似于进化上无关的噬菌体Bxb1的gp47，通过文献查阅表明，这两种RDF在趋同进化方面具有相似性。

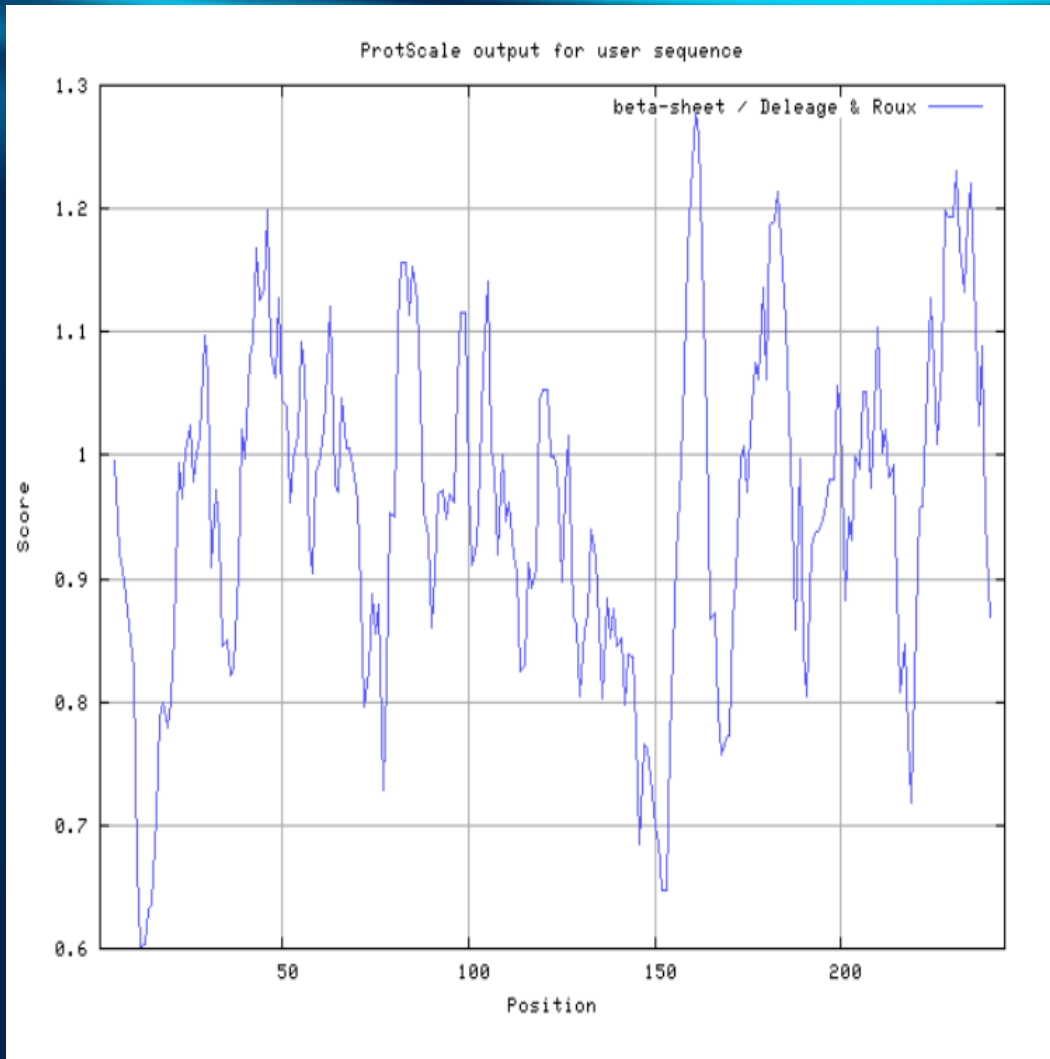
用Expasy分析gp3的氨基酸序列

LOGO



预测55-70,140-150 , 180-195,200-220位之间有alpha螺旋。

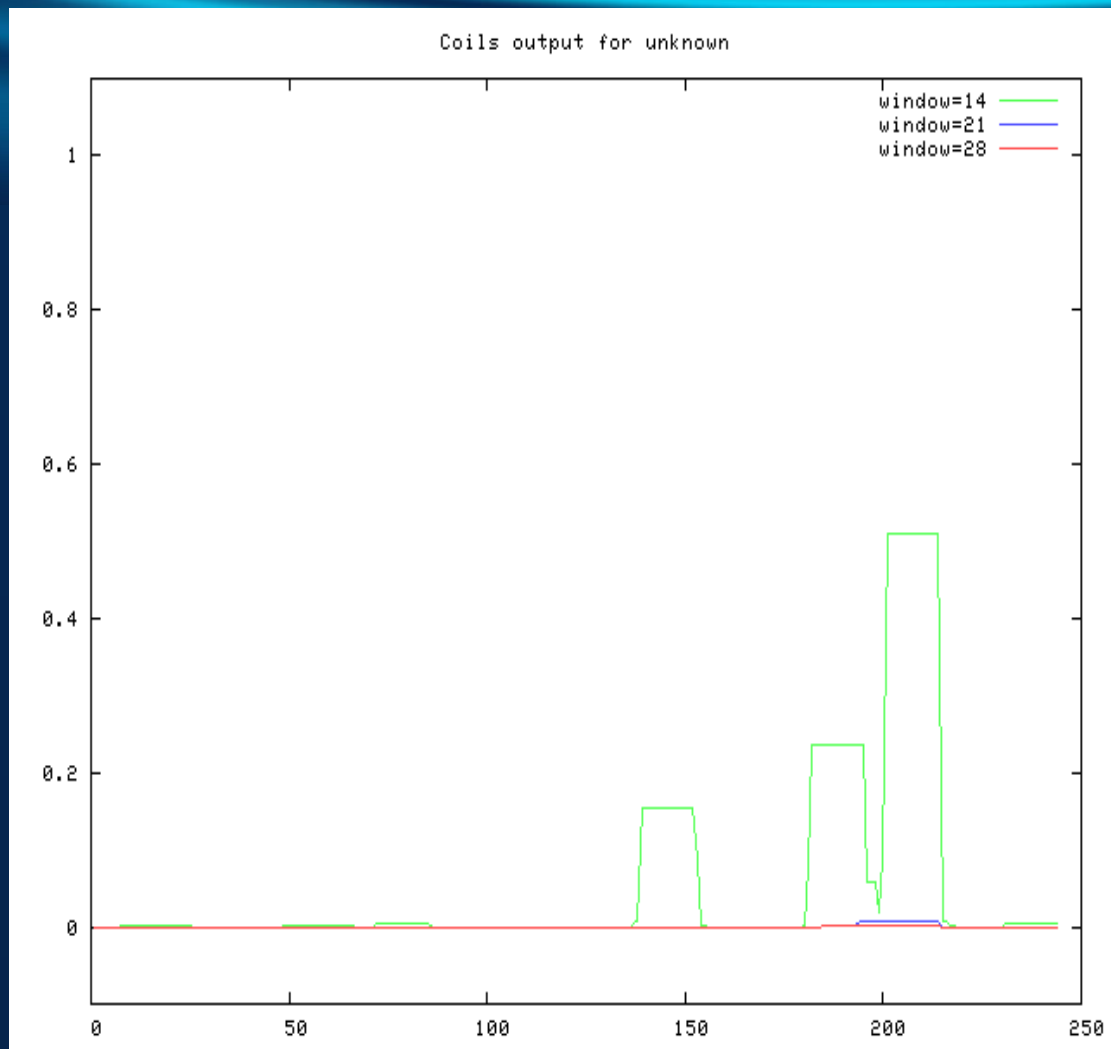




- 预测40-50,75-80,155-165,220-240位之间有 beta 折叠



用Expasy中的coils软件预测卷曲螺旋



分析在220-240位之间可能有一个卷曲螺旋。



用Weblab中的pepcoil预测卷曲螺旋

```
#####  
# Program: pepcoil  
# Rundate: Sat 9 Jun 2012 23:07:41  
# Commandline: pepcoil  
# -auto  
# -other N  
# -frame  
# -window 28  
# -coil  
# -sequence 544148  
# -outfile 544149  
# Report_format: motif  
# Report_file: 544149  
#####  
#=====  
#  
# Sequence: Gp3      from: 1    to: 244  
# HitCount: 0  
#  
# Window size: 28 residues  
#  
#=====  
Max_coil_pos at "*"   
#-----  
#-----
```

- 结果显示没有卷曲螺旋



用 SMART分析gp3的保守区

SMART SETUP FAQ ABOUT GLOSSARY WHAT'S NEW FEEDBACK keywords... Search SMART

Domains within the query sequence of 244 residues

1 100 200

low complexity region
Position: 201 to 212
E-value: N/A

Mouse over domain / undetected region for more info; click on it to go to detailed annotation; right-click to save whole protein as PNG image

Transmembrane segments as predicted by the *TMHMM2* program (■), coiled coil regions determined by the *Coils2* program (■), segments of low compositional complexity determined by the *SEG* program (■).

Signal peptides determined by the *SignalP* program (■). Hits only found by BLAST are indicated by ■ for hits in the schnipsel database and ■ for hits against PDB. Regions containing repeats detected by *Prospero*, but not covered by domains are indicated by ■. Intron positions are indicated with vertical lines showing the intron phase and exact position in AA.

You can save the results of your search for easy access in the future by bookmarking this page. It will be available for one month.

- 在201-212之间有个低复杂区域



Blast

LOGO

用gp3的氨基酸序列检索蛋白质库结果

Sequences producing significant alignments:

<u>Accession</u>	<u>Description</u>	<u>Max score</u>	<u>Total score</u>	<u>Query coverage</u>	<u>E value</u>	<u>Max ident</u>
NP_047948.1	gp3 [Streptomyces phage phiC31] >emb CAA07127.1 gp3 [Strepton	508	508	100%	0.0	100%
CAA53914.1	unnamed protein product [Streptomyces phage phiC31]	509	509	100%	0.0	100%
NP_813719.1	gp3 [Streptomyces phage phiBT1] >emb CAD80127.1 gp3 [Strepton	441	441	100%	1e-154	85%
ZP_09407037.1	hypothetical protein SPW_7341 [Streptomyces sp. W007] >gb EHM2	169	169	94%	5e-48	43%
YP_004905875.1	unnamed protein product [Kitasatospora setae KM-6054] >dbj BAJ29	109	109	76%	7e-26	38%
YP_003714756.1	gp6 [Streptomyces phage phiSASD1] >gb ADE43473.1 gp6 [Strepto	103	103	85%	2e-23	33%
ZP_04999429.1	hypothetical protein SSAG_03731 [Streptomyces sp. Mg1] >gb EDX2	94.4	94.4	43%	6e-20	45%



用gp3核苷酸序列检索蛋白质库结果：

LOGO

Sequences producing significant alignments:


<u>Accession</u>	<u>Description</u>	<u>Max score</u>	<u>Total score</u>	<u>Query coverage</u>	<u>E value</u>	<u>Max ident</u>
NP_047948.1	gp3 [Streptomyces phage phiC31] >emb CAA07127.1 gp3 [Strepton	508	508	100%	0.0	100%
CAA53914.1	unnamed protein product [Streptomyces phage phiC31]	509	509	100%	0.0	100%
NP_813719.1	gp3 [Streptomyces phage phiBT1] >emb CAD80127.1 gp3 [Strepton	441	441	100%	1e-154	85%
ZP_09407037.1	hypothetical protein SPW_7341 [Streptomyces sp. W007] >gb EHM2	169	169	94%	5e-48	43%
YP_004905875.1	unnamed protein product [Kitasatospora setae KM-6054] >dbj BAJ29	109	109	76%	7e-26	38%



对gp47所在的蛋白质家族的检索

LOGO

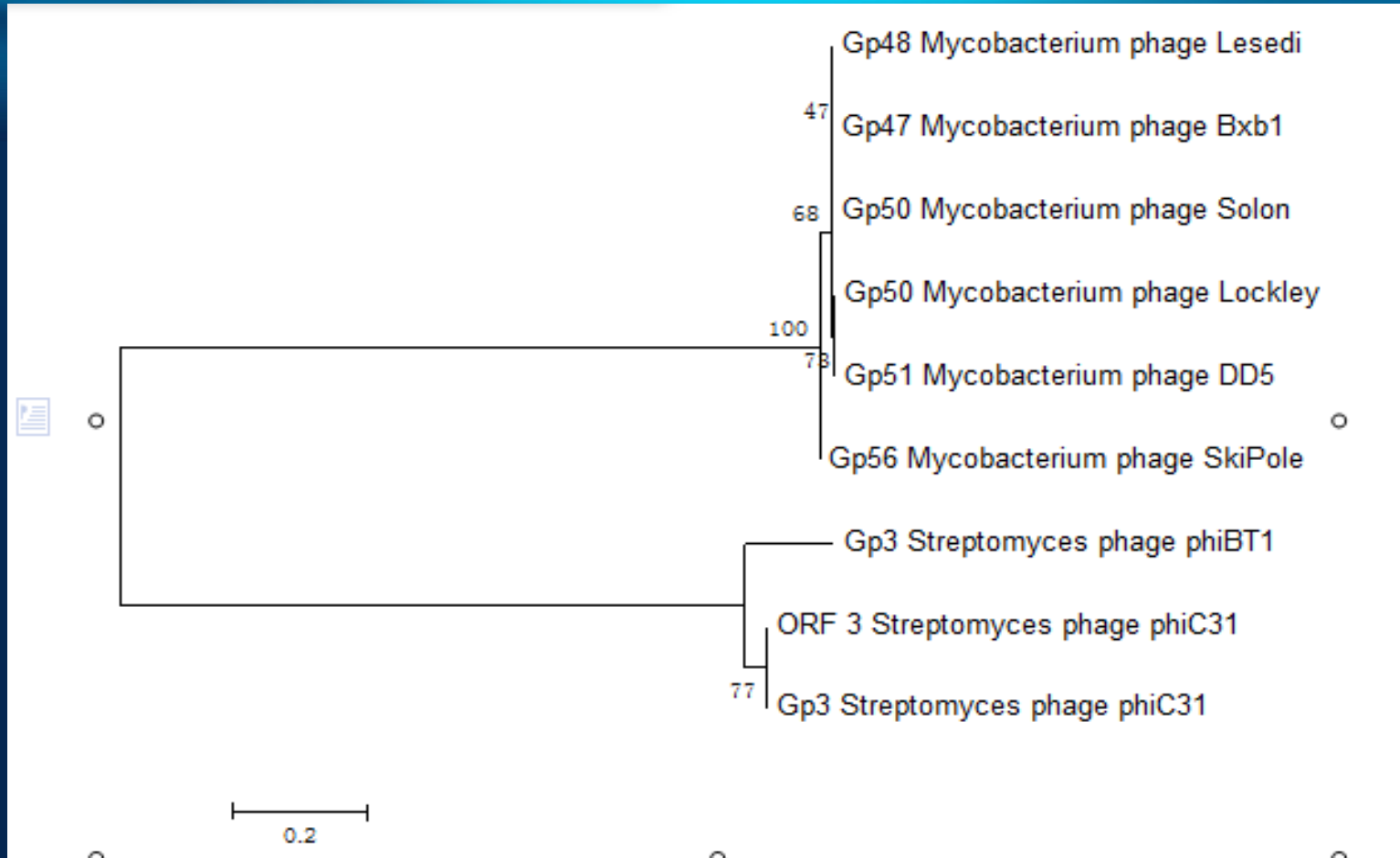
Results [Customize](#)

Cluster ID	Status	Cluster name	Size	Cluster member(s)	Organisms	Length	Identity
 UniRef90_B3VGB0	★	Cluster: Gp50	6	B3VGB0 B3VH33 B5LLQ3 G1D3J4 D2XRQ0 Q9B074	Mycobacterium phage Lockley Mycobacterium phage DD5 Mycobacterium phage Solon Mycobacterium phage Lesedi Mycobacterium phage SkiPole Mycobacterium phage Bxb1 (Mycobacteriophage Bxb1)	255	90%

进行比对发现他们保守性很强。






对gp3及Blast出的序列以及gp47蛋白质家族序列构建进化树



由于用GP3以及在Blast中得到的两条序列进行同源建模，但是都没有得到结果，而有关文献报告噬菌体Bxbl中的gp47和链球菌属噬菌体gp3蛋白功能一样，属于有趋同进化关系的两个蛋白，所以我们下面以gp47进行同源建模看看能否有预想的卷曲螺旋结构。



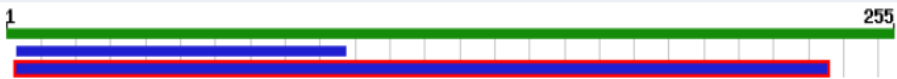
用Swiss-model同源建模




SWISS-MODEL Workspace


[Modelling](#)
[Tools](#)
[Repository](#)
[Documentation](#)


[myWorkspace] [login]


Workunit: P000003 GP47 - Overview



click on model bars

Models: [1] [2] Print/Save this page as 

Model Summary 

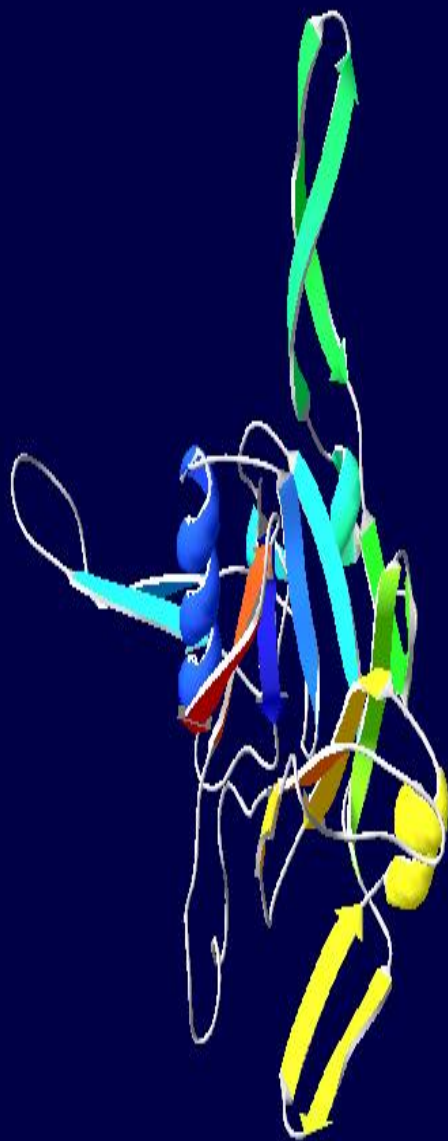
<p>Model information:</p> <p>Modelled residue range: 4 to 236</p> <p>Based on template: [1s3IB] (2.40 Å)</p> <p>Sequence Identity [%]: 14.35</p> <p>Evalue: 7.80e-10</p>	<p>Quaternary structure information: [details] ▾</p> <p>Template (1s3I): MONOMER</p> <p>Model built: SINGLE CHAIN</p>
<p>Quality information: [details] ▾</p> <p>QMEAN Z-Score: -7.43 </p>	<p>Ligand information: [details] ▾</p> <p>Ligands in the template: PO4: 1.</p> <p>Ligands in the model: none.</p>

Warning: Low QMEAN Z-scores! Only membrane proteins or models of poor quality are expected to reach such low scores.

[logs:](#) [Templates] ▾ [Alignment] ▾ [Modelling] ▾
[display model:](#) as [pdb] ▾ - as [DeepView project] ▾ - in [AstexViewer] ▾
[download model:](#) as [pdb] ± - as [Deepview project] ± - as [text] ±

- 用GP47同源建模，得出两个结果，其中model1是4-98位氨基酸，model2的是4-236位，故以model2为模板。





分析有4个
alpha螺旋，
14个beta
折叠，没
有卷曲螺
旋。



gp3氨基酸序列:

>tr|Q9T216|Q9T216_BPPHC Gp3

MAKRSIWAGDEDNKPKKRETYADDTVGRFHSGYSETNERGKVVPVALDKWRIST
GEQSVADAVAQLFGGTPVENEESTSENFIDVFTDRPKSPWIEADGIHWDMLWL
NGKLLHHCDGFDFVSHADEEMIGQPCGCPKLFDERKAAAKEYDAPNPAITVTFTL
ADDPELGRFKFQTGSWTLFKVLHEAEDDVERVGKGGAVLANLELELVEYTPKRG
PMRNKLVSYKPTITVLKSYNDIAID

gp47氨基酸序列:

>tr|Q9B074|Q9B074_BPMB1 Gp47

MTQRIVFLPDTQLPFEARKEMQAVIRFIGDVQPYGVVHIGDVLDLPQPSRWNRG
KGEFEGSVYRDADYAKKNLMEPLRKVYDGGWIGMHEGNHDLRAREYLAKNAPAL
EGTHAFDIDVLLDFDGGFVVELLPDFYDIAPGWISTHGHMGKMTLSQIAGSTALNG
AKKFGKSVVCGHTHRQAVVSHSFGYGGSVRKTVTGMEVGHLMMDMKKANYLKG
GAGNWQMGFGMLTVDGKHVKAIEIVPILGGKFTVDGQVWEV



gp3核苷酸序列

LOGO

- >gi|40807285:23098-23832 Streptomyces phage phiC31, complete genome
- ATGGCGAAGCGTTCGATCTGGGCCGGAGACGAAGACAACAAGC
CGAAGAAGCGTGAGACGTACGCCGACGACACCGTTGGGCGTTTC
CACTCCGGCTACAGCGAGACGAATGAGCGGGGGCAAGGTCGTTC
CCGTGCGGCTCGACAAGTGGCGTATCTCGACCGGCGAACAGTCC
GTTGCGGACGCTGTGGCTCAGCTCTTCGGCGGCACCCCGGTTGA
GAACGAAGAGAGCACGTCTGAGA ACTTCATCGACGTGTTACCG
ACCGGCCGAAGTCCCCGTGGATCATTGAGGCTGACGGTATCCAC
TGGGATATGAAGCTTTGGCTGAACGGCAAGCTGAAGCACCACTG
CGACGGCTTTGACTTCGTGTCGCACGCCGACGAAGAGATGATCG
GTCAGCCGTGCGGGTGCCCCGAAGCTCTTCGATGAGCGGAAGGC
AGCGGCGAAGGAGTACGACGCGCCGAACCCGGCAATCACCGTG
ACCTTCACGCTCGCCGACGACCCGGAGCTTGGCCGCTTCAAGTT
CCAGACCGGTTCTTGGACGCTCTTCAAGGTTCTGCACGAAGCCG
AAGACGACGTTGAGCGCGTCGGCAAGGGTGGCGCTGTGCTCGC
CAATCTCGAACTTGAGCTTGTGGAGTACACGCCGAAGCGTGGCC
CGATGCGGAACAAGCTTGTGAGCTACTACAAGCCGACCATCACG
GTTCTGAAGTCGTACAACGACGCGATTGCCGACTAG



gp47核苷酸序列：

LOGO

- >gi|12657854:36020-36787 Mycobacterium phage Bxb1, complete genome
- TCAGACTTCCCAGACCTGGCCGTCAACGGTGA ACTTGCCTCCCAGGATCG
GGACGATCTCAGCCTTGACATGCTTGCCGTCGACCGTGAGCATCCCGAA
GCCCATCTGCCAGTTCCCGGCTCCGCCCTTTAGATAGTTGGCCTTCTTCAT
GTCCATCAGGTGCCCGACTTCCATGCCGGTGACGGTCTTGCGCACCGAG
CCGCCGTACCCGAACGAGTGCGAGACGACAGCCTGCCGGTGCGTGTGG
CCGCAGACCACGGACTTGCCGA ACTTCTTGGCACCGTTGAGCGCTGTCG
ATCCGGCGATCTGGGATAGCGTCATCTTGCCCATGTGCCCGTGAGTGGA
GATCCAGCCCGGAGCGATGTCGTAGAAGTCAGGCAGCAGCTCCACACCG
AACCCGTCGAAGTCGAGCAGCACGTCGATGTCGAAAGCGTGCGTACCCT
CCAGGGCCGGTGCGTTCTTGGCCAGGTA CTGCGGGGCTCGCAGATCGTG
GTTGCCCTCGTG CATCCCGATCCAGCCGTCGTAGACCTTGCGCAGCGGCT
CCATCAGGTTCTTCTTGGCGTAGTCCGCGTCGCGGTACACCGAGCCCTCG
AACTCGCCCTTGGTTCCCCGGTTC CAGCGCGAGGGCTGCGGCAGGTCTA
GGACGTCACCGATATGTACCACGCCGTACGGCTGGACATCCCCGATGAA
GCGGATGACCGCTTGCATCTCTTTGCGCGCCTCGAAAGGCAACTGAGTAT
CGGGTAGAAAGACGATACGCTGAGTCAT



gp3和gp47氨基酸以及核苷酸的比对结果

LOGO

氨基酸比对结果:

Pairwise Alignment Result

LENGTH	SCORE	IDENTITY	SIMILARITY	GAPS
447	23.0	13/447 (2.9%)	20/447 (4.5%)	395/447 (88.4%)

核苷酸比对结果:

Pairwise Alignment Result

LENGTH	SCORE	IDENTITY	SIMILARITY	GAPS
847	1930.5	423/847 (49.9%)	423/847 (49.9%)	191/847 (22.6%)



gp3和gp47氨基酸以及核苷酸的比对结果

LOGO

氨基酸比对结果:

Pairwise Alignment Result

LENGTH	SCORE	IDENTITY	SIMILARITY	GAPS
447	23.0	13/447 (2.9%)	20/447 (4.5%)	395/447 (88.4%)

核苷酸比对结果:

Pairwise Alignment Result

LENGTH	SCORE	IDENTITY	SIMILARITY	GAPS
847	1930.5	423/847 (49.9%)	423/847 (49.9%)	191/847 (22.6%)

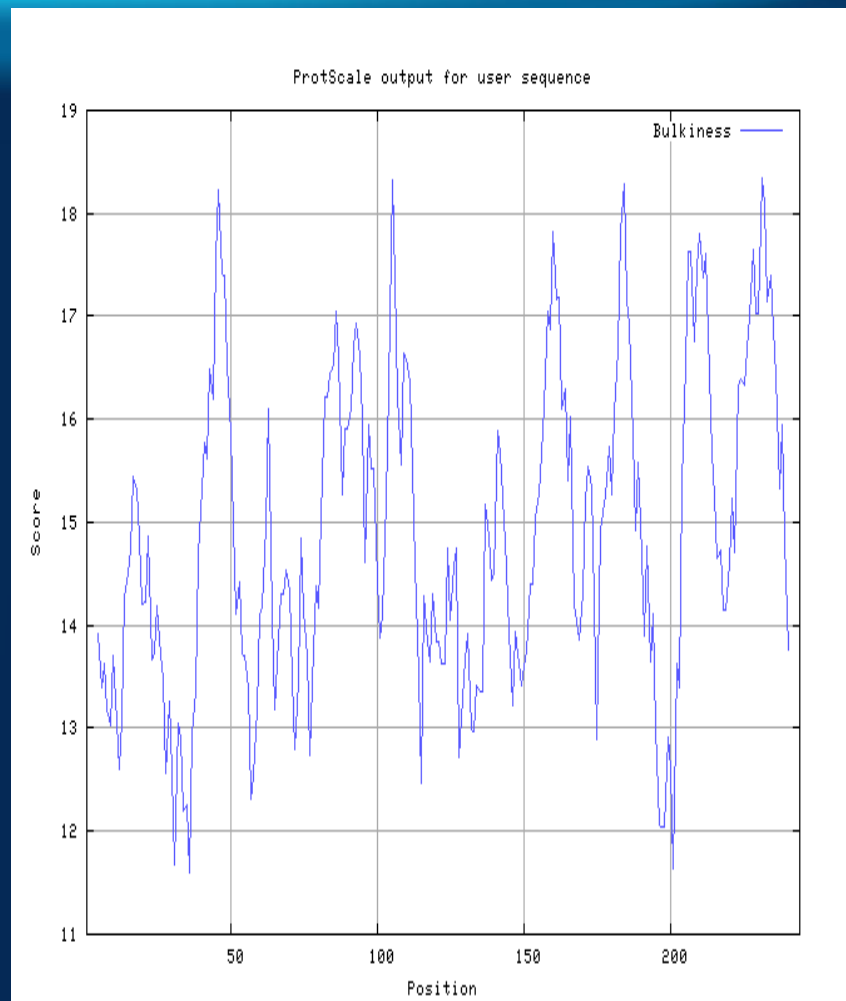
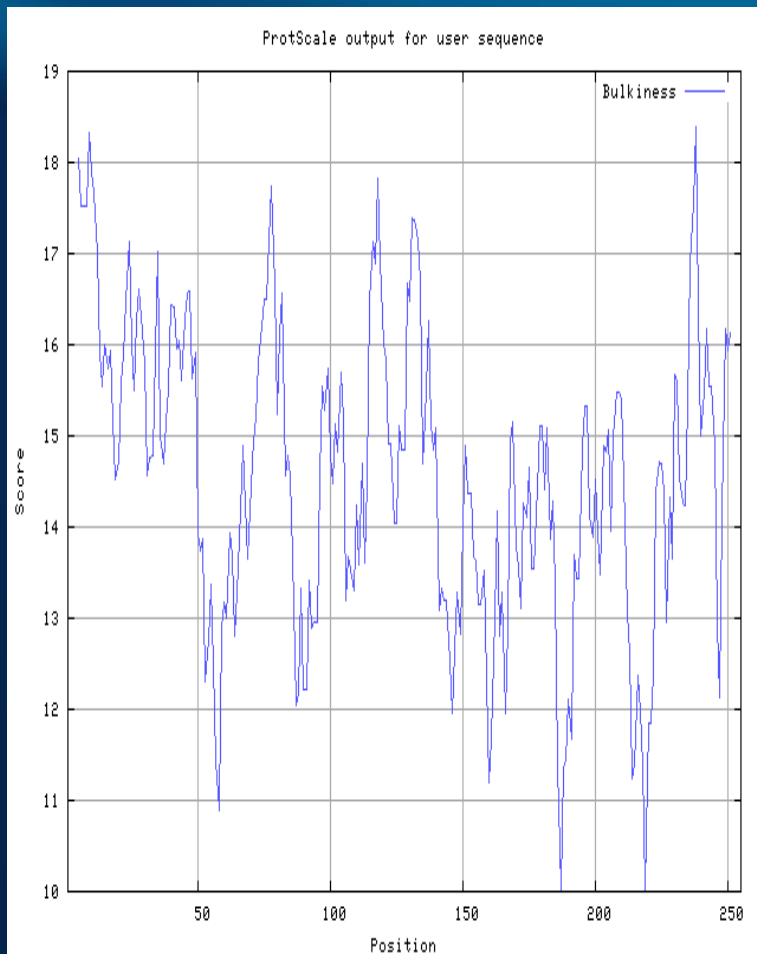


空间位阻比较

LOGO

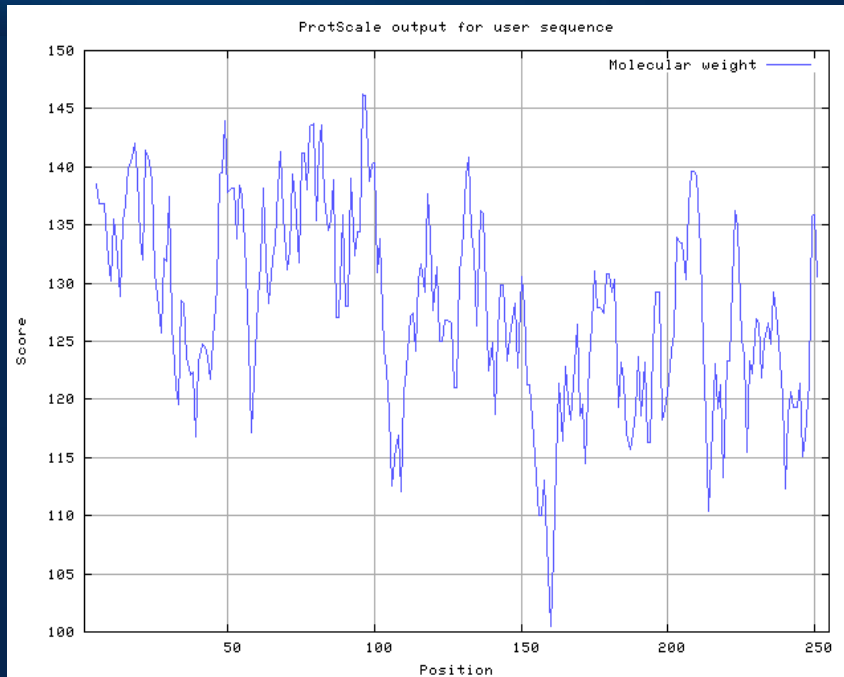
Gp47

Gp3

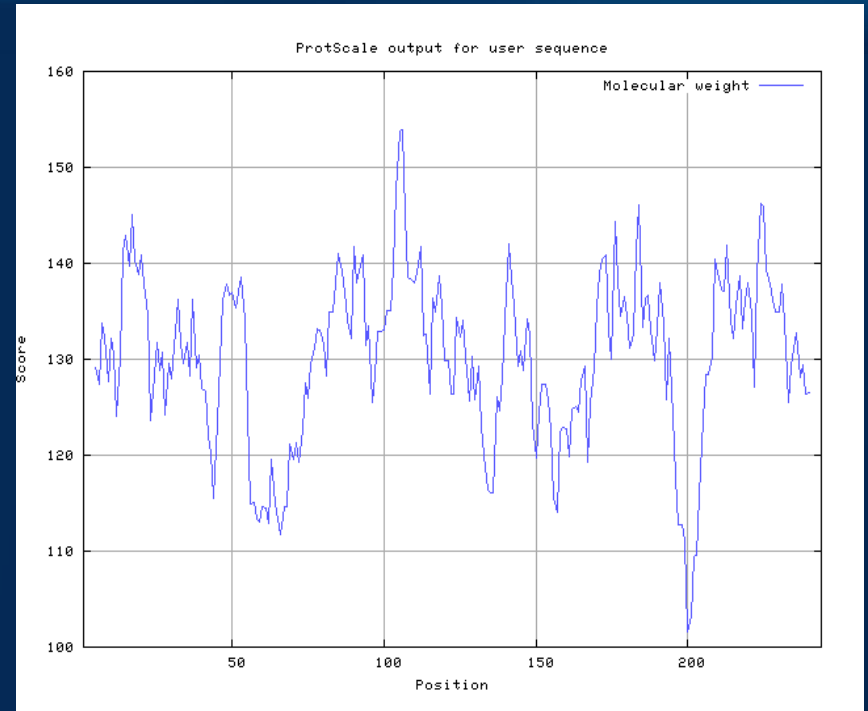


分子质量比较

Gp47

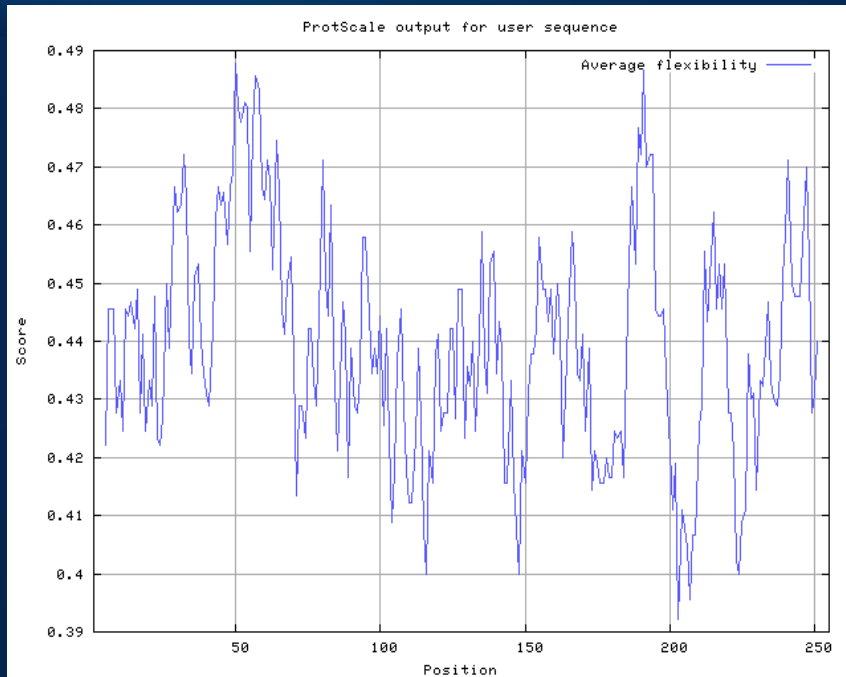


Gp3

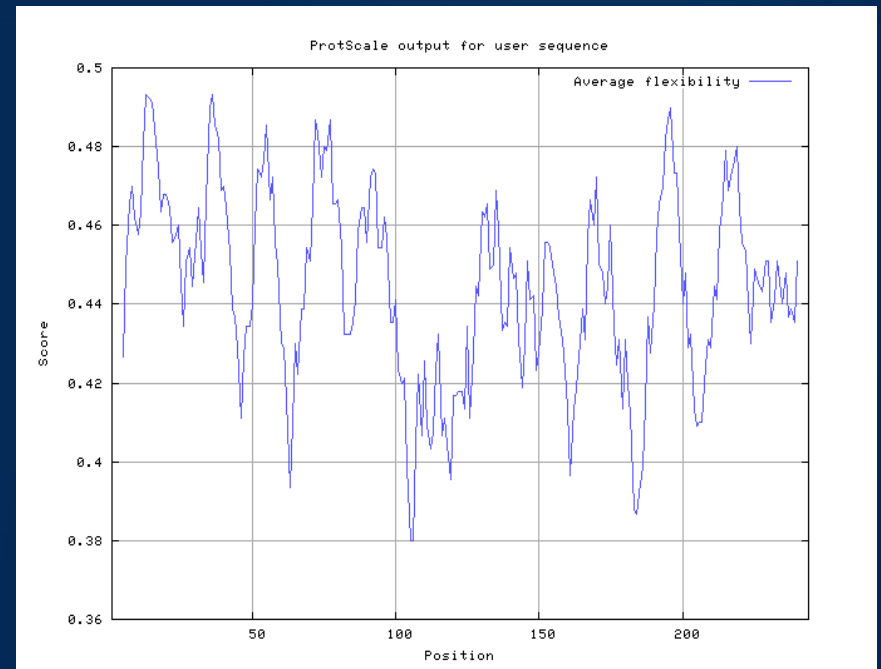


平均柔性性比较

Gp47



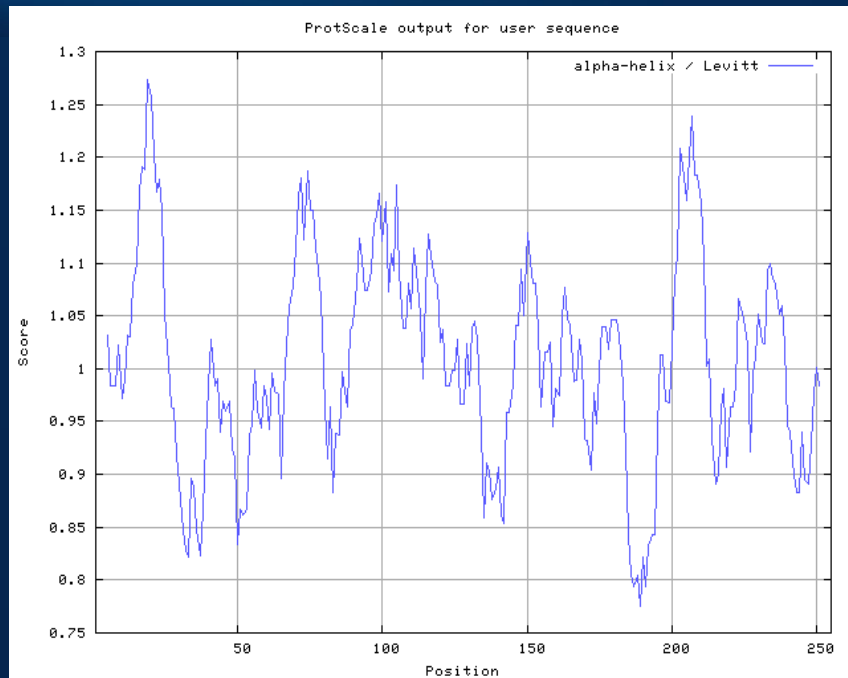
Gp3



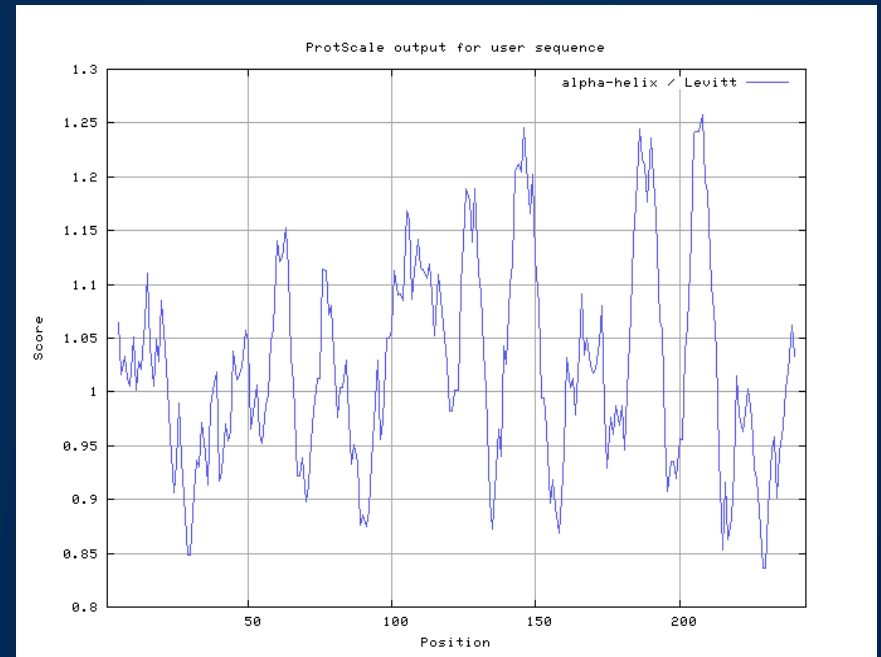
alpha螺旋比较

LOGO

Gp47

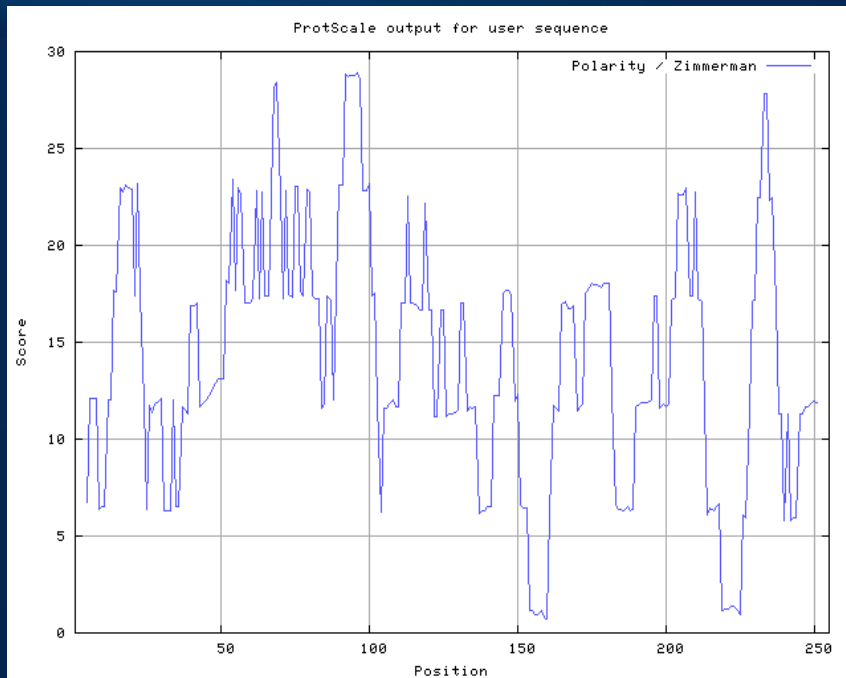


Gp3

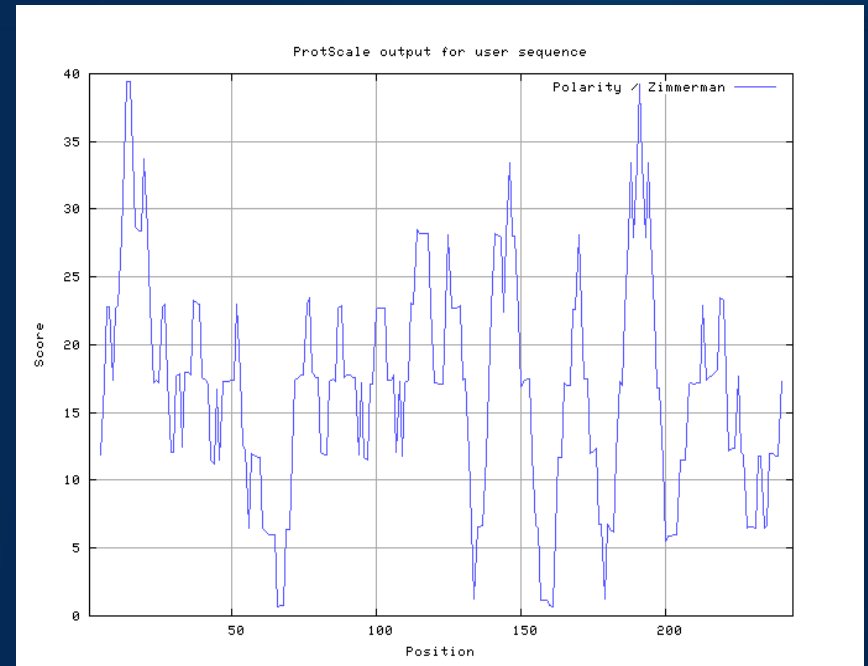


极性比较

Gp47

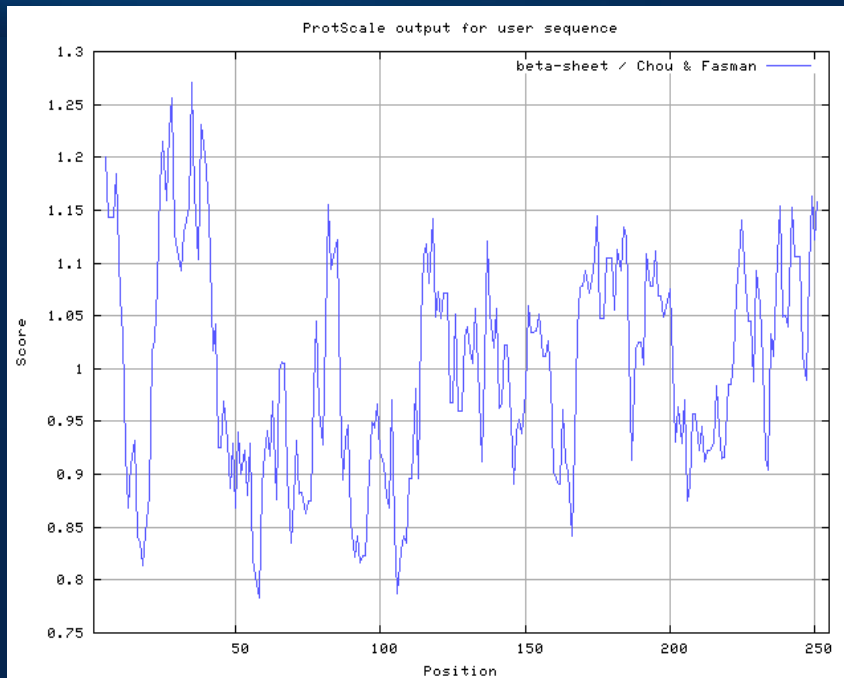


Gp3

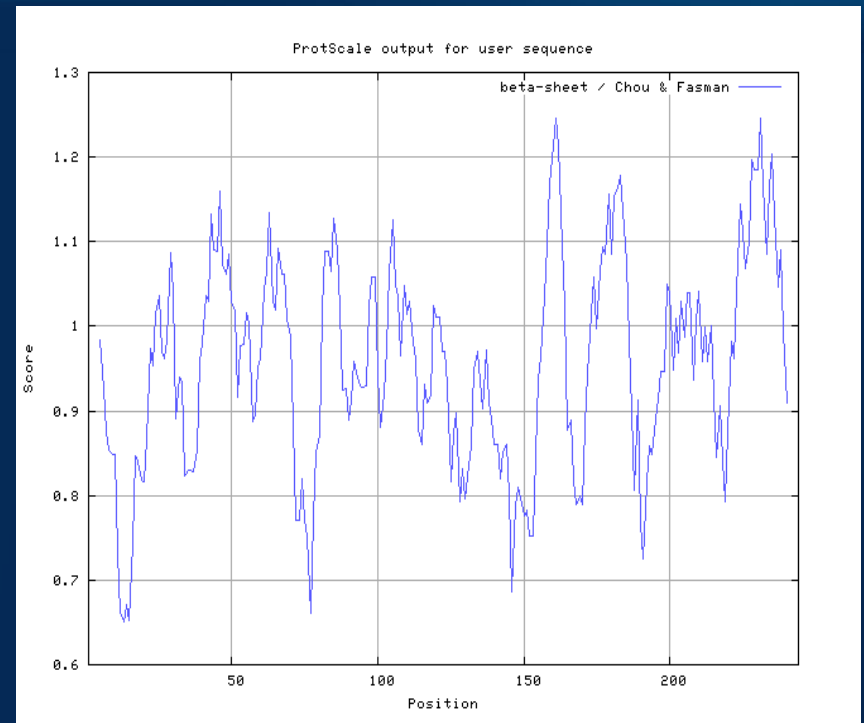


beta折叠的比较

Gp47



Gp3



结论

对gp3氨基酸结构的分析我们可以预测gp3中含有201-212位有一个低复杂区，没有很强有力的证据证明含有卷曲螺旋。

对gp3以及与它同源的三级结构的预测失败了，但对qp47的是三级结构预测得到了结果，想可以借助gp47的结构对qp3的结构进行预测。

对gp3和gp47进行的比对结果显示尽管两者的功能相似，但不论是在氨基酸序列、核酸序列、还是氨基酸结构性质的分析比较，差别显著，应该通过其他思路或方法对gp3进行预测。



- Khaleel T, Younger E, McEwan AR, Varghese AS, Smith MC. A phage protein that binds ϕ C31 integrase to switch its directionality. *Mol Microbiol.* 2011 Jun;80(6):1450-63. doi: 10.1111/j.1365-2958.2011.07696.x. Epub 2011 May 25.
- Thanafez Khaleel, Ellen Younger, Andrew R. McEwan, Anpu S. Varghese and Margaret C. M. Smith. A phage protein that binds ϕ C31 integrase to switch its directionality. *Molecular Microbiology* (2011) 80(6), 1450–1463
- Ghosh P, Wasil LR, Hatfull GF. Control of phage Bxb1 excision by a novel recombination directionality factor

致谢

LOGO

感谢罗老师在这一学期中对我们的教育与指导！

感谢春季班各位同学对我们的帮助！

感谢组内成员所作出的努力！

单击输入单位名称