

Computational Molecular Biology Biochem 218 – BioMedical Informatics 231 <u>http://biochem218.stanford.edu/</u>

Rapid Sequence Similarity Search



Doug Brutlag Professor Emeritus Biochemistry & Medicine (by courtesy)



Doug Brutlag 2010

Needleman-Wunsch Sequence Alignment

X	220	230	240	250	Х
F-	SGGNTHIYMNHVE)CKEILRREP	KELCELVISG	LPYKFRYLSTI	KE-QLK-Y
	: :: : :		:::	::::	:: ::
GDF:	IHTLGDAHIYLNHIEI	PLKIQLQREP	RPFPKLRILR	KVEKIDDFKAH	EDFQIEGYN
X	260	270	280	290	X

$$Score = \sum_{\text{Re gion}_Start}^{\text{Re gion}_End} Similarity_Weights - \sum_{\text{Re gion}_start}^{\text{Re gion}_End} Gap_Penalties$$
where:

Gap_Penalty = Gap_Start_Penalty + (Gap_Size - 1) * Gap_Size_Penalty



Needleman-Wunsch Alignment Algorithm Trace Back





Smith-Waterman Algorithm





Computer Time and Space Requirements

- Needleman-Wunsch
 - O(N*M) time and O(N*M) space
- Smith-Waterman
 - O(N*M) time and O(N*M) space





Gotoh's Improvement

		Previous Column	Current Column
			VG(i - 2, j)
Previous Row		S(i - 1, j - 2)	S(i - 1, j)
Current Row	HG(i, j - 2)	S(i, j - 1)	s(i,j)
	S(i,j) :	$=Max\begin{cases}S(i-1,j-1)+\\S(i-1,j)-GF\\S(i,j-1)-GF\\VG(i-2,j)-G\\HG(i,j-2)-\end{cases}$	s(i, j), , , , , , , , , , , , , , , , , , ,
		$\prod_{i=1}^{n} (i, j-2)^{-1}$	

s(*i*,*j*) = **S**(*i*,*j*) = S(*i*-1,*j*-1) = S(*i*,*j*-1) = S(*i*-1,*j*) = VG(*i*-2,*j*) = HG(i,j-2)= GP = **GEP**

Dayhoff score for amino acids *i* and *j* accumulated maximum score at location *i*, *j* accumulated maximum score at location *i*-1, *j*-1
 accumulated maximum score at location *i*, *j*-1
 accumulated maximum score at location *i*-1, *j* accumulated score of gap extending to *i*-1, *j* accumulated score of gap extending to *i*, *j*-1
 Gap Penalty
 Gap Extension Penalty



Computer Time and Space Requirements

- Needleman-Wunsch
 - O(N*M) time and O(N*M) space
- Smith-Waterman
 - O(N*M) time and O(N*M) space
- Gotoh improvement of Smith-Waterman
 - O(N*M) time and O(N) space
 - Remembers maximum score and its x,y location
 - Must regenerate matrix for alignment
- Myers and Miller (using Hirschberg's method)
 - O(N*M) time and O(N) space
 - Builds optimal alignment





Smith-Waterman Homology Search

Quer	:у:	HU-NS	Si Max	kima	l Score:	452	2			
PAM	Matrix:	200	Gap	Pena	alty:	5	Gap	Extension:	0.5	
No.	Score	Match	Length	DB	ID		Descripti	on	Pred. No	•
1	452	100.0	90	2	DBHB_ECOI	J	DNA-BINDI	NG PROTEIN	H 8.74e-86	
2	451	99.8	90	2	DBHB_SAL	'Y	DNA-BINDI	NG PROTEIN	H 1.54e-85	
3	336	74.3	90	2	DBHA_ECOI	J	DNA-BINDI	NG PROTEIN	H 1.64e-57	
4	336	74.3	90	2	DBHA SALI	Y	DNA-BINDI	NG PROTEIN	H 1.64e-57	
5	328	72.6	90	2	DBH_BACSI	2	DNA-BINDI	NG PROTEIN	I 1.35e-55	
6	328	72.6	92	2	DBH_BACSU	J	DNA-BINDI	NG PROTEIN	I 1.35e-55	
7	327	72.3	90	2	DBH_VIBPH	ર	DNA-BINDI	NG PROTEIN	H 2.35e-55	
8	302	66.8	90	2	DBH_PSEAE	2	DNA-BINDI	NG PROTEIN	H 2.14e-49	
9	273	60.4	91	2	DBH1_RHII	E	DNA-BINDI	NG PROTEIN	H 1.47e-42	
10	272	60.2	91	2	DBH_CLOPA	L	DNA-BINDI	NG PROTEIN	H 2.52e-42	
11	263	58.2	90	2	DBH_RHIM	2	DNA-BINDI	NG PROTEIN	H 3.18e-40	
12	261	57.7	91	2	DBH5_RHII	E	DNA-BINDI	NG PROTEIN	H 9.29e-40	
13	250	55.3	94	2	DBH_ANASH	?	DNA-BINDI	NG PROTEIN	H 3.32e-37	
14	233	51.5	93	2	DBH_CRYPH	I	DNA-BINDI	NG PROTEIN	H 2.70e-33	
15	226	50.0	95	2	DBH_THETH	I	DNA-BINDI	NG PROTEIN	I 1.07e-31	
16	210	46.5	99	3	IHFA_SERM	í A	INTEGRATI	ON HOST FAC	T 4.46e-28	
17	206	45.6	100	3	IHFA_RHOO	A	INTEGRATI	ON HOST FAC	T 3.52e-27	
18	205	45.4	99	3	IHFA_SALT	Y?	INTEGRATI	ON HOST FAC	T 5.90e-27	
19	204	45.1	99	3	IHFA_ECOI	I	INTEGRATI	ON HOST FAC	T 9.87e-27	
20	200	44.2	94	3	IHFB_ECOI	I	INTEGRATI	ON HOST FAC	T 7.71e-26	
21	200	44.2	94	3	IHFB_SERM	í A	INTEGRATI	ON HOST FAC	T 7.71e-26	
22	165	36.5	99	5	TF1_BPSP1	L	TRANSCRIP	TION FACTOR	3.42e-18	
23	147	32.5	90	2	DBH THEAC		DNA-BINDI	NG PROTEIN	H 2.12e-14	
24	76	16.8	477	2	GLGA ECOI	J	GLYCOGEN	SYNTHASE (E	C 3.80e-01	

Steps in FASTA Method Lipman & Pearson, Science 1985





FASTA Word Search (Query Hashing)



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Steps in FASTA Method







Joining Diagonals of Similarity





Steps in FastA Method





FastA Search (cont.) (HU versus SwissProt)

Alignment of hu to HLIK ASFB7

SCORES Init1: 59 Initn: 59 Opt: 84 score: 200.4 E(58800): 0.00014 Smith-Waterman score: 84; 30.2% identity in 96 aa overlap

10 20 30 40 49 hu MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVAL---VGFGT ::|::| : :|| ::::|| | : : : : :||::::| : HLIK ASFB7 MSTKKKPTITKQELYSLVAADTQLNKALIERIFTSQQKIIQNALKHNQEVIIPPGIKFTV 10 20 30 40 50 60 50 60 70 80 90 FAVKERAARTGRNPQTGKEITIAAA---KVPSFRAGKALKDAVN Hu :|| : || |:|| ||: | | | | | : ::|| | : | :| HLIK ASFB7 VTVKAKPARQGHNPATGEPIQIKAKPEHKAVKIRALKPVHDMLN 80 70 90 100





Original BLAST Algorithm Altschul et al. J. Mol. Biol. 1990 215, 403-410.

- Basic Local Alignment Search Tool
- Indexes words in database
- Calculates "neighborhood" of each word in query using BLOSUM matrix and probability threshold
- Looks up all words and neighbors from query in database index to find High-scoring Segment Pairs (HSPs)
- Extends High-scoring Segment Pairs (HSPs) left and right to maximal length
- Finds Maximal Segment Pairs (MSPs) between query and database
- Does not permit gaps in alignments





Expectation of High-scoring Segment Pairs (HSPs) Karlin and Altschul PNAS 1990, 87, 2264-2268.



Extreme Value Distribution of Scores





Original BLAST vs Smith & Waterman (Metr vs Swiss-Prot: 60 members expected)

		Penaltie	S	Thresho	old (5% exp	pectation)
			Gap	Number	Number	Number
Program	PAM	Gap	Size	Right	Wrong	Missed
				(TP/60)	(FP)	(FN/60)
5 & W	1	20	5	3	Λ	57
5 & W	50	20	5	5 97	4	22
	50	20	5	21		33
5 & W	100	20	5	42	1	18
S & W	150	20	5	51	0	9
S & W	200	20	5	53	0	7
S & W	250	20	5	50	0	10
S & W	200	5	5	2	0	58
S & W	200	10	5	53	2	7
S & W	200	20	5	53	0	7
S & W	200	40	5	53	0	7
S & W	200	80	5	51	0	9
BLAST	2	8	8	2	0	58
BLAST	50	00	00	23	0	37
BLAST	100	00	00	32	0	28
BLAST	150	00	00	35	0	25
BLAST	200	00	00	40	0	20
BLAST	250	00	00	35	0	25





cDNA Queries Require Affine Gap Penalties







Detecting Genomic Sequences with cDNA Queries



GAPPED BLAST Starts with a Two Hit Approach





GAPPED BLAST Extension of Two Hit HSP







Region Explored by GAPPED BLAST





GAPPED BLAST Extension of Two Hit HSP







CC

GAPPED BLAST Alignment

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Leghemoglobin	43	FSF	LKD	SAGV	VDSI	PKLG	АНА	EKVI	GMVF	DSA	/QLI	RAI	GEV	TV	LDGK	DGS	** ** *		90
		F	L +	V	+ +1	PK+ .	AH	+KV			L	÷	GE	V	LD	G+			
Beta globin	45	FGD	LSN	PGAV	MGNE	PKVK	AHG	KKV			LI	HSF	GEG	VHE	ILDNL	KGT	FA	LSE	90
-																			
Leghemoglobin	91	IHI	QKG	VLDP	-HF\	ννκ	EAL	LKT]	IKEAS	GDKV	ISEI	ELS	AAN	IEVA	YDGL	ATA	I 1	L40	
• -		+H	K	+DP	+F	++	L	.+ -	F	G 1	++. 1	EL	A++	-+-	G+	A A	+		
Beta globin	91	LHC	DKL	HVDP	enfi	RLLG	NVL	VVVI	LARHF	GKDE	TPI	ELC	ASY	QKV	VAGV	'ANA	L]	L 41	





Extreme Value Distribution of Scores



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Gapped BLAST Advanced Settings http://www.ncbi.nlm.nih.gov/BLAST/

- -G Cost to open gap [Integer]
 - default = 5 for nucleotides 11 proteins
- -E Cost to extend gap [Integer]
 - default = 2 nucleotides 1 proteins
- -q Penalty for nucleotide mismatch [Integer]
 - default = -3
- -r reward for nucleotide match [Integer]
 - default = 1
- -e expect value [Real]
 - default = 10
- -W wordsize [Integer]
 - default = 11 nucleotides 3 proteins





PSI-BLAST Alignment





Dynamic Programming





Generalized Dynamic Programming



) ()

PSI-BLAST Alignment I

15 VFLKTELSFALVNRKPVVPGHVLVCPLRPVERFHDLRPDEVADLF Histidine triad protein 59 L+ P V+R +L ++ DL ALV + P + ++TE 213 IVVETEHWIALVPYWAIWPFETLLLPKTHVKRLTELSDEQSKDLA 257 Uridylyltransferase 60 QTTQRVGTVVEKHFHGT-SLTFSMQDGPEAGQTVKH--VHVHVLP 101 Histidine triad protein P G+ +H +H H P +++ T + F + + 258 VILKKLTTKYDNLFETSFPYSMGFHAAPFNGEDNEHWQLHAHFYP 302 Uridylyltransferase

Histidine triad protein102 R--KAGDFHRNDSIYEELQKHDKEDFPASWRSEEEMAAEAAALRV144+++YE L ++++ AE AA R+Uridylyltransferase303 PLLRSATVRKFMVGYEMLGEN------QRDLTAEQAAERL336





Sequence Profile

Position	Probe 189 - 214 130 - 159 8 - 37	A	c	D	E	F	G	н	I	ĸ	F	Profi ∗	le N	P	Q	R	S	T	v	W	Y	Gap Opening	Gap Extension
123456789011234567890112345678901233456789011234567890333333333333333333333333333333333333	V V C D H D G S C D K S F T T K S N T L S L K R H I S K V H *	$15 \\ 16 \\ 11 \\ 12 \\ 17 \\ -5 \\ 15 \\ 32 \\ 16 \\ 67 \\ 32 \\ 9 \\ 32 \\ 19 \\ 9 \\ 15 \\ 9 \\ 13 \\ 10 \\ 5 \\ 8 \\ 7 \\ 6 \\ -1 \\ 7 \\ 9 \\ 0 \\ 6 \\ 17 \\ 9 \\ 9 \\ -4 \\ 11 \\ 10 \\ 10 \\ 10 \\ 10 \\ 10 \\ 10 \\ 1$	$\begin{array}{r} -3 \\ -8 \\ -16 \\ -6 \\ 20 \\ -3 \\ 142 \\ -13 \\ -7 \\ -15 \\ -3 \\ 142 \\ -13 \\ -17 \\ 3 \\ 1 \\ -58 \\ -10 \\ -58 \\ -18 \\ -10 \\ -58 \\ -18 \\ -10 \\ -27 \\ -7 \\ 2 \\ -17 \\ 19 \\ \end{array}$	1222223139-236-4825940435-484099-46119379533-3215281177-44013	$\begin{array}{c} 13\\ 24\\ 27\\ 13\\ 8\\ -19\\ 8\\ -49\\ 22\\ 11\\ 37\\ -30\\ 5\\ -49\\ 31\\ 9\\ 8\\ -34\\ 10\\ 9\\ 13\\ 6\\ 7\\ 5\\ 4\\ 3\\ -17\\ 16\\ 11\\ 28\\ 2\\ 18\\ 9\\ -2\\ 39\\ 14 \end{array}$	$\begin{array}{r} -8\\ -14\\ -16\\ -15\\ -57\\ -2\\ -10\\ -24\\ -4\\ -26\\ -31\\ 0\\ -25\\ -15\\ -13\\ 83\\ -9\\ -3\\ -20\\ -11\\ -25\\ -13\\ 83\\ -9\\ -3\\ -20\\ -11\\ -2\\ -1\\ 3\\ 4\\ 77\\ -22\\ -16\\ -7\\ 6\\ -11\\ -8\\ 10\\ -12\\ 14\end{array}$	$11 \\ 22 \\ 7 \\ 5 \\ 11 \\ -21 \\ 8 \\ 19 \\ 15 \\ 25 \\ 62 \\ 6 \\ 19 \\ 10 \\ 5 \\ 25 \\ 2 \\ 28 \\ 31 \\ 0 \\ 12 \\ -33 \\ 14 \\ 10 \\ 5 \\ 5 \\ 2 \\ 2 \\ 28 \\ 3 \\ -7 \\ 1 \\ 13 \\ 5 \\ 3 \\ -13 \\ 12 \\ 12 \\ 13 \\ 13$	$\begin{array}{r} 4\\ 8\\ 22\\ 8\\ 9\\ 10\\ 4\\ -7\\ 12\\ 21\\ 14\\ 3\\ 4\\ -7\\ 12\\ 11\\ 7\\ 3\\ 4\\ 7\\ 9\\ 10\\ 16\\ 5\\ 5\\ 7\\ -10\\ 14\\ 18\\ 104\\ 5\\ 12\\ 7\\ 1\\ 152\\ 21\end{array}$	$\begin{array}{c} 10 \\ 4 \\ 0 \\ 4 \\ 6 \\ 20 \\ 29 \\ 0 \\ 3 \\ 1 \\ -7 \\ 9 \\ 29 \\ -1 \\ 2 \\ 2 \\ 41 \\ 10 \\ 9 \\ 1 \\ 4 \\ 0 \\ 6 \\ 8 \\ 6 \\ 9 \\ -1 \\ 12 \\ 4 \\ 8 \\ 23 \\ -2 \\ 2 \\ 2 \\ 2 \\ 2 \\ 2 \\ 2 \\ 2 \\ 2 \\ $	$12 \\ 8 \\ 15 \\ 31 \\ -20 \\ 3 \\ -51 \\ 12 \\ 6 \\ 5 \\ -51 \\ 12 \\ 6 \\ 5 \\ -51 \\ 135 \\ 16 \\ -35 \\ 11 \\ 42 \\ 9 \\ 14 \\ 5 \\ 4 \\ 3 \\ -17 \\ 39 \\ 24 \\ 11 \\ 19 \\ 15 \\ 25 \\ 15 \\ 15 \\ 15 \\ 15 \\ 15 \\ 15$	$\begin{array}{c} 2\\ -1\\ -1\\ 0\\ 3\\ 29\\ 14\\ -73\\ -8\\ 2\\ -8\\ 2\\ -73\\ -16\\ 5\\ -73\\ -17\\ -2\\ 4\\ -1\\ 4\\ 9\\ 5\\ 7\\ -6\\ -8\\ 10\\ 2\\ 17\\ -16\end{array}$	$\begin{array}{c} 6\\ 2\\ 3\\ 11\\ -53\\ -9\\ -53\\ -5\\ 12\\ 27\\ 5\\ 10\\ 3\\ 15\\ 8\\ 5\\ 9\\ 9\\ -15\\ 8\\ 4\\ 10\\ 13\\ -27\\ 2\end{array}$	$\begin{array}{c} 11\\ 15\\ 1-3\\ -25\\ -25\\ 19\\ 24\\ 26\\ -25\\ 27\\ 14\\ 13\\ -19\\ 15\\ 11\\ 23\\ 8\\ 6\\ 7\\ 9\\ 18\\ 5\\ 3\\ 5\\ 18\\ 12\\ 53\\ 7\end{array}$	9 8 28 -24 17 9 21 6 22 14 5 9 9 8 13 -36 2 8 11 4 3 2 6 9 12 16 9 12 16 11 4 3 2 16 9 12 16 16 16 16 16 16 16 16 16 16 16 16 16	$\begin{array}{c} 7 \\ 16 \\ 315 \\ 11 \\ -2 \\ 63 \\ 24 \\ 14 \\ 15 \\ 216 \\ 88 \\ 79 \\ 55 \\ 44 \\ 217 \\ 53 \\ 219 \\ -4 \\ 73 \\ 7 \\ 7 \\ 7 \\ 7 \\ 7 \\ 7 \\ 7 \\ 7 \\ $	$ \begin{array}{r} 6 \\ 3 \\ 14 \\ 212 \\ -18 \\ -27 \\ 16 \\ -57 \\ 300 \\ -25 \\ 75 \\ 117 \\ 55 \\ 49 \\ 333 \\ 35 \\ 8 \\ 15 \\ -50 \\ 16 \\ 16 \\ 17 \\ 55 \\ 49 \\ 333 \\ 35 \\ 8 \\ 15 \\ -10 \\ 16 \\ 16 \\ 17 \\ 55 \\ 49 \\ 333 \\ 35 \\ 8 \\ 15 \\ -50 \\ 16 \\ 16 \\ 16 \\ 17 \\ 55 \\ 49 \\ 333 \\ 35 \\ 38 \\ 15 \\ -10 \\ 16 \\ 10 \\ $	14 15 8 17 -5 27 8 20 11 27 8 20 12 9 7 7 20 12 9 7 7 20 12 9 7 7 20 12 9 7 7 20 13 5 4 7 20 12 5 12 9 7 7 20 13 5 4 7 20 12 5 12 9 7 12 12 9 12 12 12 12 12 12 12 12 12 12 12 12 12	$\begin{array}{c} 32\\ 13\\ 7\\ 15\\ 12\\ -5\\ 16\\ 28\\ 15\\ 7\\ 16\\ 28\\ 28\\ 15\\ 10\\ 12\\ 33\\ 22\\ 13\\ 16\\ 11\\ 6\\ -3\\ 14\\ 8\\ -2\\ 6\\ 11\\ 120\\ -7\\ 15\\ \end{array}$	12 90 5 13 8 25 30 2 2 30 2 2 30 10 30 4 3 4 16 210 3 3 -17 8 6 10 0 5 10 3 4 -110 5 10 3 4 -110 5 10 3 2 -110 5 10 5 10 5 10 5 10 5 10 5 10 5 10	$\begin{array}{c} -22\\ -30\\ -19\\ -8\\ -21\\ 38\\ -27\\ -129\\ -26\\ -32\\ -59\\ -129\\ -38\\ -17\\ -14\\ -10\\ -6\\ 29\\ -7\\ -5\\ 20\\ 0\\ 105\\ -6\\ -15\\ -12\\ -14\\ 2\end{array}$	$\begin{array}{c} -8\\ -12\\ -11\\ -15\\ 58\\ -8\\ 101\\ -18\\ 15\\ -28\\ 101\\ -16\\ -14\\ -9\\ -6\\ -15\\ 2\\ 3\\ 0\\ 1\\ 5\\ 18\\ -13\\ 15\\ -10\\ 25\\ 9\end{array}$	25 25 25 100 100 100 100 100 24 24 24 24 24 24 24 24 24 24 24 20 100 100 100 100 100 100 100 100 100	25 25 100 100 100 100 100 100 100 100 100 10















Saccelrys



FastA Protein

http://seqweb.stanford.edu:81/gcg-bin/analysis.cgi?program=fasta-prot

Search a peptide da	atabase.				
Input sequence:	Select Fro	om: Default	Project Local	File Clipboa	rd Databa
Sequence	Descriptio	n	Туре	Length	Range
Refresh	ID KPC1_DROME Reviewed; 6	79 AA.	P	679	1 679 Clea
Input Parameters:					
Search Set		genpept T	ranslated GenBank		•
Word size		2 🛟			
List scores until E() n	eaches	10.0	(range 0.0 thru	20.0)	
Number of processor	r <u>s to use</u>	1			
Scoring Matrix		blosum50	+		
Set gap creation pen	alty	12			
Set gap extension pe	enalty	2			
Use scoring matrix to	o calculate initial diagonal scores				
Search only the top s	strand of nucleotide sequences				
Search only sequence	es entered after [m.yy]				
Only search sequence	es equal to or longer than	1	(range 1 thru 1	00000)	
Only search sequence	es equal to or shorter than	350000	(range 1 thru 3	50000)	
	list (regardless of E() value)		(range 1 thru 1	000)	
Number of scores to	list (regardless of E() value)		(lange I und I	000)	







SeqWeb FASTA http://seqweb.stanford.edu:81/gcg-bin/analysis.cgi?program=fasta-prot

Search a peptide da	atabase.				
Input sequence:	Select Fro	om: Default 🗘 P	roject Local	File Clipboa	ard Data
Sequence	Descriptio	n	Туре	Length	Rang
PKS1 DROMEL	ID KPC1_DROME Reviewed; 6	79 AA.	Р	679	1 679
Refresh					Cle
Input Parameters:					
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Word size		2 🛟			
List scores until E() r	eaches	10.0 (ra	nge 0.0 thru	20.0)	
Number of processo	rs to use	1			
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Set gap creation per	alty	12			
Set gap extension pe	enalty	2			
Use scoring matrix to	o calculate initial diagonal scores				
Search only the top	strand of nucleotide sequences				
bourten only the top					-
Search only sequence	es entered after [m.yy]				
	ac aqual to an langar than	1 (ra	nge 1 thru 1	00000)	
Only search sequence	es equal to or longer than				
Only search sequence	es equal to or shorter than	350000 (ra	nge 1 thru 3	50000)	
Only search sequence Only search sequence Number of scores to	es equal to or shorter than list (regardless of E() value)	350000 (ra.	nge 1 thru 3 nge 1 thru 1	50000) 000)	









http://seqweb.stanford.edu:81/gcg-bin/analysis.cgi?program=blastp

SLASI	-	-	-		
			<u></u>	Δ	1
	10		~		•



Peptide query against a peptide database (BLASTP).

Input sequence:	Select From: Default + Project	ocal File Clipboard Database
Sequence	Description	Type Length Range
PKS1 DROMEL	ID KPC1_DROME Reviewed; 679 AA.	P 679 <u>1679</u>
Refresh		Clear
Input Parameters:		
Search Set		(*) *
Tonone latter that and		10.0
chance alone	gnt occur more than now many times by	(range 0.0 thru
		1000.0)
Number of process	ors to use for the search	1
Filter input sequend	ces for low complex / repeat regions	
Protein scoring ma	trix	BLOSUM62
Create gapped alig	nments	
Maximum number	of sequences listed in the output	500
		(range 1 thru 1000)
Run Reset		



Saccelrys



SeqWeb BLASTP

http://seqweb.stanford.edu:81/gcg-bin/analysis.cgi?program=blastp

Input sequence:	Select From: Default + Project Lo	cal File Clipboard Da	atabase
Sequence	Description	Type Length R	ange
PKS1_DROMEL	ID KPC1_DROME Reviewed; 679 AA.	P 679 <u>1</u> .	. 679
Refresh		(Clear
Ignore hits that m	ight occur more than how many times by	10.0 (range 0.0 thr	u
<u>chance alone</u> Number of process	sors to use for the search	(range 0.0 thr 1000.0)	u
Filter input sequen	ces for low complex / repeat regions		
Protein scoring ma	itrix	BLOSUM62	
FIOLEIN SCOTING INA		4	
Create gapped alig	anments		



Saccelrys



SeqWeb PSI-BLAST Protein

http://seqweb.stanford.edu:81/gcg-bin/analysis.cgi?program=psiblast

BLAST

2

Position Specific Iterated BLAST of a peptide query against a peptide database (PSI-BLAST).

Input sequence:	Select From: Default + Project Lo	ocal File Clipboard Database
Sequence	Description	Type Length Range
PKS1 DROMEL	ID KPC1_DROME Reviewed; 679 AA.	P 679 <u>1679</u>
Refresh		Clear
Input Parameters:		
Search Set		- + T
PSI-BLAST inclusior	threshold	0.005
Ignore hits that mir	abt occur more than how many times by	10.0
chance alone	gne occur more than now many times by	(range 0.0 thru 1000.0)
Number of process	ors to use for the search	1;
Filter input sequenc	es for low complex / repeat regions	
Protein scoring mat	rix	BLOSUM62
Create gapped alig	nments	
Maximum number o	of sequences listed in the output	500
		(range 1 thru 1000)
Run Reset		



NCBI BLAST Home Page http://blast.ncbi.nlm.nih.gov/

BLAST finds regio	ns of similarity between biological sequences. more	Your Recent Results Ne
	New Aligning Multiple Protein Sequences? Try the COBALT Multiple Alignment Tool. Go	(6) - HU Search
BLAST Assemi	oled Genomes	(5) - HU Search
Choose a species g	enome to search, or list all genomic BLAST databases.	All Recent results
 <u>Human</u> <u>Mouse</u> <u>Rat</u> <u>Arabidopsis th</u> 	• Oryza sativa • Gallus gallus • Bos taurus • Pan troglodytes • Danio rerio • Microbes aliana • Drosophila melanogaster • Apis mellifera	News BLAST+ article in BMC Bioinformatics
Basic BLAST		A new article, BLAST+: architecture and application describes improvements f
Choose a BLAST pr	ogram to run.	other new BLAST features
nucleotide blast	Search a nucleotide database using a nucleotide query Algorithms: blastn, megablast, discontiguous megablast	EST
protein blast	Search protein database using a protein query Algorithms: blastp, psi-blast, phi-blast	
blastx	Search protein database using a translated nucleotide query	Tip of the Day
tblastn	Search translated nucleotide database using a protein query	More tips
tolastn	Search translated nucleotide database using a protein query	



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NCBI BLAST Input http://blast.ncbi.nlm.nih.gov/

BLAST Home Rece	Basic Local Alignment Search Tool ant Results Saved Strategies Help		My NCBI Welcome brutlag. [
CBI/ BLAST/ bla	stp suite		
astn blastp blas	tx tblastn tblastx		
	BLASTP programs search protein databases using a protein que	y. more	Reset page
Enter Querv	Sequence		<u>Bookmark</u>
Enter accession	number, gi, or FASTA sequence 😡 Clear	Query subrance	
>HU-NS1			7
MNKSQLIDKIA	AGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTGRNPQT	From	1
GKEIT	KALKDAVN	То]
Or, upload file	Choose File no file selected		
Job Title	HU-NS1		
	Enter a descriptive title for your BLAST search @		
□ Align two or i	more sequences 😡		
01	and Oat		
Choose Sea	rch Set		
Database	Swissprot protein sequences(swissprot) Swissprot protein sequences(swissprot)		2
Organism		+	N
Optional	Enter organism common name, binomial, or tay id. Only 20 ton tays will be shown	0	
Evaluate			
Optional	Models (XM/XP) Environmental sample sequences		
Entrez Query			
Optional	Enter an Entrez query to limit search (9)		
Drogram Or	Institut		
Program Se	IECHOT		
Algorithm	O blaste (sectors particip DI AOT)		
Algorithm	o blastp (protein-protein BLAST)		
Algorithm	 Diastp (protein-protein BLAST) O PSI-BLAST (Position-Specific Iterated BLAST) 		
Algorithm	 Diastp (protein-protein BLAST) PSI-BLAST (Position-Specific Iterated BLAST) PHI-BLAST (Pattern Hit Initiated BLAST) 		
Algorithm	 Diastp (protein-protein BLAST) PSI-BLAST (Position-Specific Iterated BLAST) PHI-BLAST (Pattern Hit Initiated BLAST) Choose a BLAST algorithm (g) 		

(cc)

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NCBI BLAST Parameters http://blast.ncbi.nlm.nih.gov/

Algorithm paramet	Note: Parameter values that differ from the default are highlighted in yellow an
ked with 🕈 sign	
General Param	neters
Max target sequences	 \$ 500 \$ Select the maximum number of aligned sequences to display
Short queries	Automatically adjust parameters for short input sequences 😡
Expect threshold	10 💿
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BLAST	Search database Swissprot protein sequences(swissprot) using Blastp protein-protein BLAST

NCBI BLAST Conserved Domains

http://blast.ncbi.nlm.nih.gov/





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Search for similar domain architectures

2 Refine search

List of domain hits			?
Description	Pssmld	Multi-dom	E-value
+]cd00591, HU_IHF, Integration host factor (IHF) and HU are small heterodimeric members of the DNABII	29683	no	2e-17
(+) PRK10664, PRK10664, transcriptional regulator HU subunit beta; Provisional	138067	no	6e-23
(+) pfam00216, Bac_DNA_binding, Bacterial DNA-binding protein	109281	no	1e-21
(+)smart00411, BHL, bacterial (prokaryotic) histone like domain	128689	no	8e-21
[+]COG0776, HimA, Bacterial nucleoid DNA-binding protein [DNA replication, recombination, and repair]	31119	no	8e-19
(+) PRK10753, PRK10753, transcriptional regulator HU subunit alpha; Provisional	138142	no	2e-16
(+) PRK00199, ihfB, integration host factor subunit beta; Reviewed	134166	no	1e-12
(+) TIGR00988, hip, integration host factor, beta subunit	130061	no	1e-11
(+) PRK00285, ihfA, integration host factor subunit alpha; Reviewed	134205	no	9e-11
(+)TIGR00987, himA, integration host factor, alpha subunit	130060	no	6e-10



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1P78 guer	_B v	1 MNKGE	LVD A VAE LID K IAA	KA.[3]	. KKQADA	VLTAALET	IIEAVSSG VTESLKEG	DKVTLVG	FGSFESR	ERKARE ERAARI	GRNPK	TNEKM TGKEI	HEIPAT
1P78 quer gi 1	_B Y 4194648	1 MNKGE 1 MNKSQI 1 MNKTEI	LVDAVAE LIDKIAA LIHQVAE	GA.[3]	. KKQADA . KAAAGF . KKDAGE	VLTAALET ALDAIIAS	IIEAVSSG VTESLKEG IAESLAOG	DKVTLVG DDVALVG	FGSFESF FGTFAVK FGNFEVF	ERKARE ERAARI ERAARK	GRNPK GRNPQ GRNPQ	TNEKM TGKEJ TGELJ	MEIPAT ITIAAA IDIAAT
1P78 quer gi 1 gi 8	_B y 4194648 1857031	1 MNKGEI 1 MNKSQI 1 MNKTEI 3 LTKDQI	LVDAVAE LIDKIAA LIHQVAE LIQDIAE	KA.[3] GA.[3] RT.[3] AI.[3]	. KKQADA . KAAAGF . KKDAGE . KTTVRS	VLTAALET ALDAIIAS VVNTVFDV ALDQLAEI	IIEAVSSG /TESLKEG IAESLAQG /KDALEND	DKVTLVG DDVALVG DSVQLIG GEITLPG	FGSFESE FGTFAVE FGNFEVE IGKLKVS	ERKARE ERAARI ERAARI ERPARI	GRNPK' GRNPQ' GRNPQ'	TNEKM TGKEI TGEEI TGKAI	MEIPAT ITIAAA IDIAAT IEIAAK
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NCBI Blast Distance Tree

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Tree view for RID: NWZ0ANPY016, query ID: lcl/82136, database: swissprot





NCBI Blast Distance Tree

http://blast.ncbi.nlm.nih.gov/ Tree view for RID: NWZOANPY016, query ID: lcl/82136, database: swissp

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## BLAST High Scores http://blast.ncbi.nlm.nih.gov/

### Distribution of 100 Blast Hits on the Query Sequence @

Mouse over to see the defline, click to show alignments





## BLAST High Scores http://blast.ncbi.nlm.nih.gov/

			Score	E
	Sequences producing si	gnificant alignments:	(Bits)	Value
	sp POACF6.1 DBHB ECO57	RecName: Full=DNA-binding protein HU-b	174	1e-43
	sp POA1R8.1 DBHB SALTY	RecName: Full=DNA-binding protein HU-b	172	3e-43
	sp P52681.1 DBHB SERMA	RecName: Full=DNA-binding protein HU-b	158	8e-39
	sp P05384.3 DBHB PSEAE	RecName: Full=DNA-binding protein HU-beta	142	7e-34
	sp Q9KQS9.1 DBHB VIBCH	RecName: Full=DNA-binding protein HU-beta	135	5e-32
	SP Q9KHS6.1 DBHB PSEF5	RecName: Full=DNA-binding protein HU-beta	129	5e-30 G
	sp Q9LA96.2 DBHA AERHY	RecName: Full=DNA-binding protein HU-a	127	1e-29
	sp P52680.1 DBHA SERMA	RecName: Full=DNA-binding protein HU-a	124	1e-28
	sp POACF2.1 DBHA ECO57	RecName: Full=DNA-binding protein HU-a	123	3e-28
	sp POA1R6.1 DBHA SALTY	RecName: Full=DNA-binding protein HU-a	122	7e-28
	sp Q87E48.1 DBH XYLFT	RecName: Full=DNA-binding protein HU	121	8e-28 G
	sp P64389.1 DBHB NEIMB	RecName: Full=DNA-binding protein HU-b	121	1e-27
	sp P28080.1 DBHA VIBPR	RecName: Full=DNA-binding protein HU-a	119	4e-27
	sp Q9PE38.1 DBH_XYLFA	RecName: Full=DNA-binding protein HU	116	4e-26
	sp Q5HFV0.1 DBH_STAAC	RecName: Full=DNA-binding protein HU >s	_114	1e-25 G
1	sp P43722.1 DBH_HAEIN	RecName: Full=DNA-binding protein HU	114	2e-25
	sp POA3H0.1 DBH_BACST	RecName: Full=DNA-binding protein HU; A	112	7e-25
	sp Q9KV83.1 DBHA_VIBCH	RecName: Full=DNA-binding protein HU-a	_111	9e-25
	sp Q9KDA5.1 DBH1_BACHD	RecName: Full=DNA-binding protein HU-1	110	2e-24
	sp Q8KA69.1 DBH_BUCAP	RecName: Full=DNA-binding protein HU	109	4e-24
	sp P08821.2 DBH1_BACSU	RecName: Full=DNA-binding protein HU 1	108	8e-24
	sp P57144.1 DBH_BUCAI	RecName: Full=DNA-binding protein HU	108	1e-23
	sp Q9CK94.1 DBH_PASMU	RecName: Full=DNA-binding protein HU	108	1e-23
	sp Q9JR30.1 DBHC_NEIMA	RecName: Full=DNA-binding protein HU-b	107	1e-23
	sp Q9K7K5.1 DBH2_BACHD	RecName: Full=DNA-binding protein HU-1	107	2e-23
	sp P96045.1 DBH_STRTR	RecName: Full=DNA-binding protein HU	104	1e-22
	sp POA3I0.1 DBH_STRP3	RecName: Full=DNA-binding protein HU >s	102	4e-22
	sp Q9XB21.1 DBH STRMU	RecName: Full=DNA-binding protein HU	102	6e-22
	SP 09XB22.1 DBH STRDO	RecName: Full=DNA-binding protein HU	100	3e-21
	SP P68573.1 DBHZ BACSU	RecName: Full=SPBC2 prophage-derived D	99.0	7e-21
	SP DOS295 1 DBH STRGN	RecName: Full=DNA-binding protein HU	98.2	1e-20
	SPIPUSSOS.I DBH CLOPA	RecName: Full-DNA-binding protein HU	90.5	40-20
	SP 099822 1 DBHA BUCPP	RecName: Full=DNA-binding protein HU	95.9	5e-20
	SP 09CT64 1 DBH LACLA	RecName: Full=DNA-binding protein HU	93.9	20-19
	ap D02344 2 DBH PHIME	RecName: Full=DNA-binding protein HPm	92 1	60-19
	SD 09HTLO 1 DBHA DEFAF	RecName: Full=DNA-binding protein HU-a	89 7	40-19
	SD P02348 1 DBH5 BHTTE	RecName: Full=DNA-binding protein HDL5	86 3	40-17
	SD 068X.16 1 DBH BTCTT	RecName: Full=DNA-binding protein HU	85 5	80-17
	SP D05514 2 DBH ANACD	RecName: Full=DNA-binding protein HU	84 3	20-16
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Doug Brutlag 2010

## BLAST Low Scores http://blast.ncbi.nlm.nih.gov/

GENE ID: 1107533 asr3935 | DNA binding protein HU [Nostoc sp. PCC 7120] (10 or fewer PubMed links)

Score = 84.3 bits (207), Expect = 2e-16, Method: Compositional matrix adjust Identities = 39/89 (43%), Positives = 59/89 (66%), Gaps = 0/89 (0%)

Query 1 MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTG 60 MNK +L+D +A A ++K A L A + ++ E++ GD V LVGFG+F +ER AR G Sbjct 1 MNKGELVDAVAEKASVTKKQADAVLTAALETIIEAVSSGDKVTLVGFGSFESRERKAREG 60

Query 61 RNPQTGKEITIAAAKVPSFRAGKALKDAV 89 RNP+T +++ I A +VP+F AGK ++ V Sbjct 61 RNPKTNEKMEIPATRVPAFSAGKLFREKV 89

> <u>sp P29214.1 DBH_GUITH</u> C RecName: Full=DNA-binding protein HU homolog; AltNa protein Length=93

GENE ID: 857075 hlp | DNA-binding protein hu homolog [Guillardia theta] (10 or fewer PubMed links)

Score = 84.0 bits (206), Expect = 2e-16, Method: Compositional matrix adjust Identities = 36/90 (40%), Positives = 59/90 (65%), Gaps = 0/90 (0%)

Query1MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTG60MNKSQLIKIASK+ + +++ + +++ + G+ VLVGFG+F +ER AR GSbjct1MNKSQLISKIAYYTKYSKTDIEKIITSMLEIIVDTVATGEKVTLVGFGSFEARERKAREG60

Query 61 RNPQTGKEITIAAAKVPSFRAGKALKDAVN 90 RNP+TG+++ + A+++P+F G ++ VN Sbjct 61 RNPRTGEKLFLPASRIPTFSVGNFFRNKVN 90

> <u>sp P36206.2 DBH_THEMA</u> RecName: Full=DNA-binding protein HU Length=90

Score = 84.0 bits (206), Expect = 2e-16, Method: Compositional matrix adjust Identities = 44/90 (48%), Positives = 59/90 (65%), Gaps = 0/90 (0%)

 Query
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 MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTG
 60

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 LD I+ ++TE+L +G+ V +VGFG+F V++ AAR G

 Sbjct
 1
 MTKKELIDRVAKKAGAKKKDVKLILDTILETITEALAKGEKVQIVGFGSFEVRKAAARKG
 60

Query 61 RNPQTGKEITIAAAKVPSFRAGKALKDAVN 90 NPQT K ITI KVP F+ GKALK+ V Sbjct 61 VNPQTRKPITIPERKVPKFKPGKALKEKVK 90



## BLAST Typical Alignments http://blast.ncbi.nlm.nih.gov/

GENE ID: 1107533 asr3935 | DNA binding protein HU [Nostoc sp. PCC 7120] (10 or fewer PubMed links)

Score = 84.3 bits (207), Expect = 2e-16, Method: Compositional matrix adjust Identities = 39/89 (43%), Positives = 59/89 (66%), Gaps = 0/89 (0%)

Query 1 MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTG 60 MNK +L+D +A A ++K A L A + ++ E++ GD V LVGFG+F +ER AR G Sbjct 1 MNKGELVDAVAEKASVTKKQADAVLTAALETIIEAVSSGDKVTLVGFGSFESRERKAREG 60

```
Query 61 RNPQTGKEITIAAAKVPSFRAGKALKDAV 89
RNP+T +++ I A +VP+F AGK ++ V
Sbjct 61 RNPKTNEKMEIPATRVPAFSAGKLFREKV 89
```

> <u>sp P29214.1 DBH_GUITH</u> C RecName: Full=DNA-binding protein HU homolog; AltNa protein Length=93

GENE ID: 857075 hlp | DNA-binding protein hu homolog [Guillardia theta] (10 or fewer PubMed links)

```
Score = 84.0 bits (206), Expect = 2e-16, Method: Compositional matrix adjust
Identities = 36/90 (40%), Positives = 59/90 (65%), Gaps = 0/90 (0%)
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Query 1 MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTG 60
MNKSQLI KIA SK + + +++ + +++ G+ V LVGFG+F +ER AR G
Sbjct 1 MNKSQLISKIAYYTKYSKTDIEKIITSMLEIIVDTVATGEKVTLVGFGSFEARERKAREG 60
```

```
Query 61 RNPQTGKEITIAAAKVPSFRAGKALKDAVN 90
RNP+TG+++ + A+++P+F G ++ VN
Sbjct 61 RNPRTGEKLFLPASRIPTFSVGNFFRNKVN 90
```

> <u>sp P36206.2 DBH_THEMA</u> RecName: Full=DNA-binding protein HU Length=90

```
Score = 84.0 bits (206), Expect = 2e-16, Method: Compositional matrix adjust
Identities = 44/90 (48%), Positives = 59/90 (65%), Gaps = 0/90 (0%)
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Query1MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTG60M K +LID++AAKLD I+ ++TE+L +G+ V +VGFG+F V++ AAR GSbjct1MTKKELIDRVAKKAGAKKKDVKLILDTILETITEALAKGEKVQIVGFGSFEVRKAAARKG60
```

```
Query 61 RNPQTGKEITIAAAKVPSFRAGKALKDAVN 90
NPQT K ITI KVP F+ GKALK+ V
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Sbjct 61 VNPQTRKPITIPERKVPKFKPGKALKEKVK 90
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## NCBI Blast Taxonomy Report http://www.ncbi.nlm.nih.gov/BLAST/

Tax BLAST Report

Index				
<ul> <li>Lineage Report</li> <li>Organism Report</li> <li>Taxonomy Report</li> <li>Help</li> </ul>				
Lineage Report				
<u>root</u> <u>cellular organisms</u> <u>Bacteria</u> <u>Proteobacteria</u> <u>proteobacteria</u> <u>Gammaproteobacteria</u> [ <u>o-proteobacteria</u> ] <u>c. Enterobacteriace</u> [ <u>o-proteobacteria</u> ] <u>c. Enterobacteriace</u> [ <u>o-proteobacteria</u> ]				
<u>Escherichia</u> [ <u>enterobacteria</u> ] <u>Escherichia coli</u> [ <u>enterobacteria</u> ] Escherichia coli [ <u>157:87</u>	125 2	hite	enterobacterial	DNA-binding protein HU-bets (NS1) (HU-1) >gi182581655[gp]
<u>Escherichia coli</u>	65 2	hits	[enterobacteria]	Integration host factor subunit beta (IHF-beta) >qi 67466
<u>Sernatia marcescens</u>	116 4	hits	[enterobacteria]	DNA-binding protein HU-beta (NSI) (HU-1) DNA-binding protein HU-beta (NSI) (HU-1)
Buchnera aphidicola (Acvitnosionon Disum) .	91 3	hits	[enterobacteria]	DNA-binding protein HU
<u>Salmonella typhi</u>	65 1	hita	[enterobacteria]	Integration host factor subunit beta (IHF-beta) >gi 54041
<u>Yersinia Destis</u>	63 <u>2</u> 29 1	hits	[enterobacteria]	Integration host factor subunit beta (IHF-beta)
<u>Pseudomonas aeruginosa</u>	105 4	hits	[g-proteobacteria]	DNA-binding protein HU-beta
<u>Vibito Cholefac</u> <u>Accordonas hydrophila</u>	100 1	hit	<u>g-proteobacteria</u>	DNA-binding protein HU-alpha
<u>Xylella fastidiosa Temeculal</u>	98 3	hits	<u>g-proteobacteria</u>	DNA-binding protein HU
<u>Vibrio proteolyticus</u>	93 <u>1</u>	hit	[g-proteobacteria]	DNA-binding protein HU-alpha
<u>Haemoonilus ihriuenzae</u> <u>Pasteurella multocida</u>	82 3	hits	<u>g-proteobacteria</u>	DNA-binding protein HU DNA-binding protein HU
<u>Pseudomonas putida KT2440</u>	68 <u>2</u>	hits	<u>g-proteobacteria</u>	Integration host factor subunit beta (IHF-beta) >q1/60392 Integration host factor subunit beta (IHF-beta)
<u>Mannheimia haemolytica</u>	67 <u>2</u> 65 <u>2</u>	hits hits	<u>g-proteobacteria</u> ] <u>g-proteobacteria</u> ]	Integration host factor subunit beta (IHF-beta) Integration host factor subunit beta (IHF-beta)
<u>Vibrio vulnificus</u> <u>Vibrio vulnificus YJ016</u>	65 <u>2</u> 63 <u>1</u>	hits hit	[ <u>g-proteobacteria</u> ] [ <u>g-proteobacteria</u> ]	Integration host factor subunit beta (IHF-beta) Integration host factor subunit beta (IHF-beta)
<u>Haemophilus ducrevi</u> <u>Xanthomonas campestris pv. campestris</u>	62 <u>2</u> 59 <u>2</u>	hits hits	[ <u>g-proteobacteria</u> ] [ <u>g-proteobacteria</u> ]	Integration host factor subunit beta (IHF-beta) Integration host factor subunit beta (IHF-beta)
Xanthomonas axonopodis pv. citri	58 1	hit	[g-proteobacteria]	Integration host factor subunit beta (IHF-beta)

93 2 hits [b-proteobacteria]



Neisseria meningitidis serogroup



DNA-binding protein HU-beta >gi|54041019|sp|P64388|DBHB



## NCBI Blast Organism Report http://www.ncbi.nlm.nih.gov/BLAST/

### **Organism Report**

Escherichia coli 0157:H7 [enterobacteria] taxid 83334		
gi 82581654 sp POACF6 DBHB_ECO57_DNA-binding protein HU-be	125	4e-29
gi 82581650 sp POACF2 DBHA ECO57 DNA-binding protein HU-al	95	3e-20
Salmonella typhimurium [ enterobacteria] taxid 602		
gi 60392171 sp POA1R8 DBHB SALTY DNA-binding protein HU-be	_124	1e-28
gi   60392169   sp   POA1R6   DBHA_SALTY DNA-binding protein HU-al	95	3e-20
gi 60392433 sp POA1S0 IHFA_SALTY Integration host factor s	56	2e-08
Serratia marcescens [ enterobacteria] taxid 615		
gi 1706310 sp P52681 DBHB_SERMA DNA-binding protein HU-bet	116	2e-26
gi 1706309 sp P52680 DBHA SERMA DNA-binding protein HU-alp	98	4e-21
gi 124290 sp P23303 IHFB_SERMA Integration host factor sub	63	1e-10
gi 400046 sp P23302 IHFA SERMA Integration host factor sub	57	1e-08
Pseudomonas aeruginosa [g-proteobacteria] taxid 287		
gi 12643997 sp P05384 DBHB_PSEAE DNA-binding protein HU-beta	_105	3e-23
gi 14194645 sp Q9HTL0 DBHA_PSEAE DNA-binding protein HU-alpha	76	2e-14
gi 2495249 sp Q51473 IHFB PSEAE Integration host factor su	66	2e-11
gi 2495247 sp Q51472 IHFA PSEAE Integration host factor su	56	2e-08
Vibrio cholerae [g-proteobacteria] taxid 666		
gi 14194651 sp Q9KQS9 DBHB_VIBCH_DNA-binding protein HU-beta	_104	6e-23
gi 14194652 sp Q9KV83 DBHA_VIBCH_DNA-binding protein HU-alpha	90	2e-18
gi 14194866 sp Q9KQT4 IHFB_VIBCH Integration host factor s	63	2e-10
gi 14194867 sp Q9KSN4 IHFA_VIBCH Integration host factor s	56	2e-08

Aeromonas hydrophila [g-proteobacteria] taxid 644



CC



## NCBI Blast Taxomoy Report http://www.ncbi.nlm.nih.gov/BLAST/

### **Taxonomy Report**

root	143 hits	93	orgs	
. cellular organisms	141 hits	91	orgs	
Bacteria	138 hits	88	orgs	
Proteobacteria	102 hits	55	orgs	
Gammaproteobacteria	63 hits	29	orgs	
Enterobacteriaceae	24 hits	11	orgs	[Enterobacteriales]
Escherichia	4 hits	2	orgs	
Escherichia coli	4 hits	2	orgs	
Escherichia coli 0157:H7	2 hits	1	orgs	
Salmonella	4 hits	2	orgs	
Salmonella typhimurium	3 hits	1	orgs	
Salmonella typhi	1 hits	1	orgs	
Serratia marcescens	4 hits	1	orgs	[Serratia]
Buchnera aphidicola	7 hits	3	orgs	Buchneral
Buchnera aphidicola (Acvrthosiphon pisum) .	3 hits	1	orgs	•
Buchnera aphidicola (Schizaphis graminum) .	3 hits	1	orgs	
Buchnera aphidicola (Baizongia pistaciae) .	1 hits	1	orgs	
Erwinia chrysanthemi str. 3937	2 hits	1	orgs	[Dickeva; Erwinia chrysanthemi]
Yersinia pestis	2 hits	1	orgs	[Yersinia]
Shigella flexneri	1 hits	1	orgs	Shigella
Pseudomonas	9 hits	4	orgs	[Pseudomonadales; Pseudomonadaceae]
Pseudomonas aeruginosa	4 hits	1	orgs	[Pseudomonas aeruginosa group]
Pseudomonas fluorescens Pf-5	1 hits	1	orgs	[Pseudomonas fluorescens group; Pseudomonas fluorescens]
Pseudomonas putida KT2440	2 hits	1	orgs	[ Pseudomonas putida group; Pseudomonas putida]
Pseudomonas syringae pv. tomato	2 hits	1	orgs	[ Pseudomonas syringae group; Pseudomonas syringae group genomosp
Vibrio	10 hits	5	orgs	[Vibrionales; Vibrionaceae]
Vibrio cholerae	4 hits	1	orgs	
Vibrio proteolyticus	1 hits	1	orgs	
Vibrio parahaemolyticus	2 hits	1	orgs	
Vibrio vulnificus	3 hits	2	orgs	
Vibrio vulnificus YJ016	1 hits	1	orgs	
Aeromonas hydrophila	1 hits	1	orgs	[Aeromonadales; Aeromonadaceae; Aeromonas]
Xanthomonadaceae	9 hits	4	orgs	[Xanthomonadales]
Xvlella	6 hits	2	orgs	
Xvlella fastidiosa	6 hits	2	orgs	
Xvlella fastidiosa Temeculal	3 hits	1	orgs	
		-	- 24	







## Decypher Search Engine http://decypher.stanford.edu/





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2010

**DNA to Protein Profile** 

Protein Profile to DNA

Algorithm	Query vs. Database Types		Algorithm	Query vs. Database Types	
Tera-Blast [™] N	DNA to DNA	0		DNA to DNA	
	DNA to DNA	0	Smith-Waterman	DNA to Protein	
Tera-Blast [™] P	DNA to Protein	0	Standard, Semi-Global, Double-Affine	Protein to Protein	
	Protein to DNA	0		Protein to DNA	
	Protein to Protein	0	FrameSearch	DNA to DNA	
Tera-Probe TM	DNA to DNA	0	Symmetric Frame Independent TM for DNA to	DNA to Protein	
	Genomic DNA to Coding DNA	0	DNA	Protein to DNA	
	Coding DNA to Genomic DNA	0		DNA to Protein HMM	
Dama Data atiwa TM	Genomic DNA to Protein	0	Hidden Markov Model	Protein to Protein HMM	
GeneDetective	Protein to Genomic DNA	0	(HMM)	Protein HMM to Protein	
	Genomic DNA to Protein HMM	0		Protein HMM to DNA	
	Protein HMM to Genomic DNA	0		DNA to Protein HMM	
ClustelW	DNA	0	rimm riamesearch	Protein HMM to DNA	
Ciustaiw	Protein	0		DNA to Protein Profile	
Target Build	All	0	BrofileSearch	Protein To Protein Profile	
			Promesearch	Protein Profile to Protein	
				Protein Profile to DNA	

**Profile FrameSearch** 





## Decypher Search Engine Input http://decypher.stanford.edu/

	biocomputing solut	ions DeCypher	Ð
De Pro	Cypher Smith-Waterman Sea otein Query vs. Protein Databas	e	
0	Job Description:	DeCypher Smith-Waterman Search Protein Query vs. Protein Database	
0	E-mail Address:		
7	Return Results:	To your web browser 🗘 As: Web Page	
0	Protein Query: Click <i>Browse</i> to upload your local file, or paste query data into the text box. Use only one query entry if requesting a Graphic reply. <u>Use Example Query</u>	Choose File no file selected >HU-NS1 MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTGRNPQTGKEIT IAAAKVPSFRAGKALKDAVN	
0	Protein Database: Select only one if requesting an iterated search.	Description         Entries         Symbols Updated         \$           Treponema pallidum (NCBI         1036         356216 19-March-2003         *           tumoraa         1         393 19-December-2004         *           Uniprot-SwissProt:         57.7         497293         175274719 10-September-2009         *           Uniprot-TREMBL:         57.7         9145906         2958343657 10-September-2009         *           Ureaplasma urealyticum (         614         231000 19-March-2003         *	
0		Submit	
0	Job Options:	Hide	
0	Algorithm Variation:	Local (Standard)	
3	Filter Query:	Significance Threshold: 10	
7	Max Scores:	500 (2) Weight Matrix: blosum62 \$	
0	Max Alignments:	20	
3	Identity Symbol:	Match Letter	
1	Show Significance:	E-Value	
3	Gap Open Penalty:	12	
0	Gap Extend Penalty:	2	





## Decypher Search Engine Results http://decypher.stanford.edu/

```
RANK 19 Score = 297.00 E Value = 9.2e-033
Q = CGI Temp17444106e02.seq
QF = 1 \# Q Symbols = 90
T = sp | Q87E48 | DBH XYLFT
TF = 1 \#T Symbols = 94
D = DNA-binding protein HU OS=Xylella fastidiosa (strain Temeculal / ATCC 700964) GN=hup PE=3 SV=
Identical Match = 57 Similar = 73 Total # Of Gaps = 0
Identity: Alignment = 64% Query = 63% Target = 60%
Similarity: Alignment = 82% Query = 81% Target = 77%
OS =
            1 \quad OE =
                        89 TS =
                                       1 TE =
                                                     89
        1 MNKSOLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTG
0
          MNK++LID +AA A++SK AGRA+DA++ +TE+LKEGD V LVGFGTF V++RA R G
        1 MNKTELIDGVAAAANLSKVEAGRAIDAVVNEITEALKEGDSVTLVGFGTFQVRQRAERPG
0
       61 RNPQTGKEITIAAAKVPSFRAGKALKDAV
         RNP+TG+ I IAA+ PSF+ GKALKDAV
       61 RNPKTGEPIMIAASNNPSFKPGKALKDAV
RANK 20 Score = 296.00 E Value = 1.3e-032
Q = CGI Temp17444106e02.seq
QF = 1 \# Q Symbols = 90
T = sp | P64388 | DBHB NEIMA
TF = 1 \#T Symbols = 89
D = DNA-binding protein HU-beta OS=Neisseria meningitidis serogroup A GN=hupB PE=3 SV=1
Identical Match = 62 Similar = 72 Total # Of Gaps = 0
Identity: Alignment = 69% Query = 68% Target = 69%
Similarity: Alignment = 80% Query = 80% Target = 80%
OS =
            1 \quad OE =
                        89 TS =
                                        1 TE =
                                                     89
        1 MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTFAVKERAARTG
0
         MNKS+LI+ IA ADISKAAA +ALDA
                                       +VT +LK+GD V LVGFGTF V ERA R G
т
        1 MNKSELIEAIAQEADISKAAAQKALDATTNAVTTALKQGDTVTLVGFGTFYVGERAERQG
       61 RNPOTGKEITIAAAKVPSFRAGKALKDAV
         RNP+TG+ +TIAAAK P FRAGKALKDA+
       61 RNPKTGEPLTIAAAKTPKFRAGKALKDAL
```



## Decypher Search Engine Results http://decypher.stanford.edu/

Download All Hits (For Internet Explorer E

### Its for Job CGI_Temp17444106e02 pher Smith-Waterman Search Protein Que

ERY LOCUS] HU-NS1

#### ults for: HU-NS1; (Length=93)

ERY LENGTH] 90 ARCH TYPE] AA (AMINO ACID) TRIX] d:\decypher\matrix/blos EN PENALTY] -12.00 TEND PENALTY] -2.00 ALE FACTOR] 1 Lmum possible score for this (

	SCORE	QF	TAP	RGET ACC	ESSIT	E_VALUE	E DESCRIPTION			
	432.00	1	sp	POACF6	DBHB 1	2.2e-051	DNA-binding	protein	HU-beta OS=Escherichia co	Li 广
	432.00	1	sp	POACF5	DBHB 1	2.2e-051	DNA-binding	protein	HU-beta OS=Escherichia co	Li 🕕
	432.00	1	sp	POACF4	DBHB 1	2.2e-051	DNA-binding	protein	HU-beta OS=Escherichia co	Li
	432.00	1	sp	POACF7	DBHB 1	2.2e-051	DNA-binding	protein	HU-beta OS=Shigella flexn	eri
	428.00	1	sp	POA1R9	DBHB 1	7.9e-051	DNA-binding	protein	HU-beta OS=Salmonella typ	ni (
	428.00	1	sp	POA1R8	DBHB 1	7.9e-051	DNA-binding	protein	HU-beta OS=Salmonella typ	nim
	394.00	1	sp	P52681	DBHB 1	3.8e-046	5 DNA-binding	protein	HU-beta OS=Serratia marce	sce:
	356.00	1	sp	P05384	DBHB 1	6.7e-041	DNA-binding	protein	HU-beta OS=Pseudomonas ae	cug.
	331.00	1	sp	Q9KQS9	DBHB 1	1.9e-037	7 DNA-binding	protein	HU-beta OS=Vibrio cholera	a Gi
	324.00	1	sp	Q9KHS6	DBHB 1	1.7e-036	5 DNA-binding	protein	HU-beta OS=Pseudomonas fl	lor
	311.00	1	sp	Q9LA96	DBHA 1	1.1e-034	1 DNA-binding	protein	HU-alpha OS=Aeromonas hyd	ropi
	301.00	1	sp	P52680	DBHA 1	2.6e-033	3 DNA-binding	protein	HU-alpha OS=Serratia marc	esc
	298.00	1	sp	POACF2	DBHA 1	6.7e-033	3 DNA-binding	protein	HU-alpha OS=Escherichia c	oli
	298.00	1	sp	POACF1	DBHA 1	6.7e-033	3 DNA-binding	protein	HU-alpha OS=Escherichia c	oli
	298.00	1	sp	POACFO	DBHA 1	6.7e-033	3 DNA-binding	protein	HU-alpha OS=Escherichia c	oli
	298.00	1	sp	POACF3	DBHA 1	6.7e-033	3 DNA-binding	protein	HU-alpha OS=Shigella flex	her.
	297.00	1	sp	POA1R7	DBHA 1	9.2e-033	3 DNA-binding	protein	HU-alpha OS=Salmonella ty	phi
	297.00	1	sp	POA1R6	DBHA 1	9.2e-033	3 DNA-binding	protein	HU-alpha OS=Salmonella ty	phi
	297.00	1	sp	Q87E48	DBH_:1	9.2e-033	3 DNA-binding	protein	HU OS=Xylella fastidiosa	(st:
	296.00	1	sp	P64388	DBHB 1	1.3e-032	2 DNA-binding	protein	HU-beta OS=Neisseria meni:	ngi
	296.00	1	sp	P64389	DBHB 1	1.3e-032	2 DNA-binding	protein	HU-beta OS=Neisseria meni	ngi
	295.00	1	sp	P28080	DBHA 1	1.7e-032	2 DNA-binding	protein	HU-alpha OS=Vibrio proteo	Lyt.
	284.00	1	sp	Q9PE38	DBH_1	5.7e-031	DNA-binding	protein	HU OS=Xylella fastidiosa	JN=
	283.00	1	sp	P43722	DBH_11	7.9e-031	DNA-binding	protein	HU OS=Haemophilus influen	zae
States of the local division of the local di							20 102770 00100 PC	2,240		

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(cc)





#### Job Details

#### Return to top

[BEGIN JOB STATUS]







## Decypher Search Sequence Alignments http://decypher.stanford.edu/

Home Page

Build HMM Model ) Click once!

Results for Job CGI_Temp19051203772 DeCypher Smith-Waterman Search Protein Query vs. Protein Database

#### View dendrogram

HU-NSI			
swissprot	sp	POACF6	DBHB_EC057
swissprot	sp	POACF5	DBHB_ECOL6
swissprot	sp	POACF4	DBHB_ECOLI
swissprot	sp	POACF7	DBHB_SHIFL
swissprot	sp	POA1R9	DBHB SALTI
swissprot	sp	POA1R8	DBHB SALTY
swissprot	sp	P52681	DBHB SERMA
swissprot	sp	P05384	DBHB PSEAE
swissprot	sp	Q9KQS9	DBHB VIBCH
swissprot	sp	Q9KHS6	DBHB PSEF5
swissprot	sp	Q9LA96	DBHA AERHY
swissprot	sp	P52680	DBHA SERMA
swissprot	sp	POACF2	DBHA ECO57
swissprot	sp	POACF1	DBHA ECOL6
swissprot	sp	POACFO	DBHA ECOLI
swissprot	sp	POACF3	DBHA SHIFL
swissprot	sp	POA1R7	DBHA SALTI
swissprot	sp	POA1R6	DBHA SALTY
swissprot	sp	Q87E48	DBH XYLFT
swissprot	sp	P64388	DBHB NEIMA
swissprot	sp	P64389	DBHB NEIMB
swissprot	sp	P28080	DBHA VIBPR
_			_
HU-NS1			
swissprot	sp	POACF6	DBHB EC057
swissprot	sp	POACF5	DBHB ECOL6
swissprot	sp	POACF4	DBHB ECOLI
swissprot	sp	POACF7	DBHB SHIFL
swissprot	sp	POA1R9	DBHB SALTI
swissprot	sp	POA1R8	DBHB SALTY
swissprot	sp	P52681	DBHB SERMA
swissprot	sp	P05384	DBHB PSEAE
swissprot	sp	Q9KQS9	DBHB VIBCH
swissprot	sp	Q9KHS6	DBHB PSEF5
swissprot	sp	Q9LA96	DBHA AERHY
swissprot	sp	P52680	DBHA SERMA
swissprot	sp	POACF2	DBHA ECO57
swissprot	sp	POACF1	DBHA ECOL6
swissprot	sp	POACFO	DBHA ECOLI
swissprot	sp	POACF3	DBHA SHIFL
swissprot	sp	POA1R7	DBHA SALTI
swissprot	sp	POA1R6	DBHA SALTY
swissprot	sp	Q87E48	DBH XYLFT
swissprot	sp	P64388	DBHB NEIMA
swissprot	sp	P64389	DBHB NEIMB
swissprot	sp	P28080	DBHA VIBPR

MNKSOLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTF MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTF MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTF MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTF MNKSQLIDKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTF MNKSOLIEKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTF MNKSQLIEKIAAGADISKAAAGRALDAIIASVTESLKEGDDVALVGFGTF MNKSQLIDKIAAGADISKAAAGRALDAVIASVTDSLKAGDDVALVGFGSF MNKSELIDAIAASADIPKAVAGRALDAVIESVTGALKAGDSVVLVGFGTF MNKTOLVEOIAANADISKASAGRALDAFIEAVSGTLQSGDQVALVGFGTF MNKSELIDAIAASADLPKAAAGRALDAVIESVTGALKAGDSVVLVGFGTF MNKAQLVDAIAAKADLSKAQAKVALEEIINGITQSLKEGDAVQLVGFGTF MNKTQLIDVIADKADLSKAQAKLALESTLAAITESLKEGDAVQLVGFGTF MNKTOLIDVIAEKAELSKTOAKAALESTLAAITESLKEGDAVOLVGFGTF MNKTQLIDVIAEKAELSKTQAKAALESTLAAITESLKEGDAVQLVGFGTF MNKTQLIDVIAEKAELSKTQAKAALESTLAAITESLKEGDAVQLVGFGTF MNKTQLIDVIAEKAELSKTQAKAALESTLAAITESLKEGDAVQLVGFGTF MNKTQLIDVIADKAELSKTQAKAALESTLAAITESLKEGDAVQLVGFGTF MNKTQLIDVIADKAELSKTQAKAALESTLAAITESLKEGDAVQLVGFGTF MNKTELIDGVAAAANLSKVEAGRAIDAVVNEITEALKEGDSVTLVGFGTF MNKSELIEAIAOEADISKAAAOKALDATTNAVTTALKOGDTVTLVGFGTF MNKSELIEAIAQEADISKAAAQKALDATTNAVTTALKQGDTVTLVGFGTF MNKTQLIDFIAEKADLSKAQAKAALEATLDGVTDALKEGDQVQLIGFGTF

AVKERAARTGRNPQTGKEITIAAAKVPSFRAGKALKDAVN----AVKERAARTGRNPQTGKEITIAAAKVPSFRAGKALKDAVN----AVKERAARTGRNPQTGKEITIAAAKVPSFRAGKALKDAVN----AVKERAARTGRNPQTGKEITIAAAKVPSFRAGKALKDAVN----AVKERAARTGRNPQTGKEITIAAAKVPSFRAGKALKDAVN----AVKERAARTGRNPQTGKEITIAAAKVPSFRAGKALKDAVN----AVKERAARTGRNPQTGKEITIAAAKVPSFRAGKALKDAVN-TVRERSARTGRNPOTGKEIKIAARKVPAFRAGKALKDAVN-AVKERAARTGRNPOTGKPIKIAAAKIPGFKAGKALKDAVN--SVRTRAARTGRNPKTGEEIKIAEAKVPSFKAGKALKDACN----SVTDRPARIGRNPQTGKTLEIAAAKKPGFKAGKALKEAVN----KVNHRAGRTGRNPOTGKEIOIAAANVPSFVAGKALKDAVK----KVNHRSERTGRNPQTGKEIKIAAANVPAFVSGKALKDAVK----KVNHRAERTGRNPQTGKEIKIAAANVPAFVSGKALKDAVK----KVNHRAERTGRNPQTGKEIKIAAANVPAFVSGKALKDAVK----KVNHRAERTGRNPQTGKEIKIAAANVPAFVSGKALKDAVK----KVNHRAERTGRNPQTGKEIKIAAANVPAFVSGKALKDAVK----KVNHRAERTGRNPQTGKEIKIAAANVPAFVSGKALKDAVK----KVNHRAERTGRNPQTGKEIKIAAANVPAFVSGKALKDAVK----**OVRORAERPGRNPKTGEPIMIAASNNPSFKPGKALKDAVKSSAG** YVGERAERQGRNPKTGEPLTIAAAKTPKFRAGKALKDAL-----YVGERAERQGRNPKTGEPLTIAAAKTPKFRAGKALKDAL-----KVNHRAARTGRNPKTGAEIQIAAANVPAFVAGKALKDAVK----





## Decypher Search Sequence Alignments http://decypher.stanford.edu/

### Dendrogram

### Return to top





## General DNA Similarity Search Principles

- Search both Strands
- Translate ORFs
- Use most sensitive search possible
  - UnGapped BLAST for infinite gap penalty (PCR & CHIP oligos)
  - Gapped BLAST for most searches
  - Smith Waterman or megaBLAST or discontinuous MegaBLAST for cDNA/genome comparisons
  - cDNA =>Zero gap-length penalty
  - Consider using transition matrices
  - Ensure that expected value of score is negative
- Examine results with exp. between 0.05 and 10
- Reevaluate results of borderline significance using limited query





## General Protein Similarity Search Principles

- Chose between local or global search algorithm
- Use most sensitive search algorithm available
  - Original BLAST for no gaps
  - Smith-Waterman for most flexibility
  - Gapped BLAST for well delimited regions
  - PSI-BLAST for families
  - Initially BLOSUM62 and default gap penalties
  - If no significant results, use BLOSUM30 and lower gap penalties
  - Ensure expected score is negative
- Examine results between exp. 0.05 and 10 for biological significance
- Beware of long hits or those with unusual amino acid composition
- Reevaluate results of borderline significance using limited query





## SeqWeb v_{3.1}

Saccelrys

# SeqWeb Comparison Programs

http://seqweb.stanford.edu:81/gcg-bin/programs.cgi?name=comparison

## SeqWeb v_{3.1}

Saccelrys

•							
	Programs	Managers		Help Topics   Support			
Programs Comparison	Comparison						
Database Searching Similarity	Use these programs <b>BestFit</b> Makes an opti by inserting of	to compare two or m mal alignment of the aps to maximize the r	bore sequences. best segment of similarity between two sequences. Optimal sources of matches using the local homology algorithm of Sm	alignments are found hith and Waterman.			
Reference	Locally align two nucleic acid sequences. <u>Locally align two peptide sequences.</u>						
Mapping	ClustalW+ Creates a multiple alignment by progressively adding sequences to an alignment.						
Pattern Recognition	Align several nucleic acid sequences.						
Primer Selection Protein Analysis	Compare Compares two similar.	peptide or nucleic ac	id sequences and creates a graph that shows where the two	sequences are			
Nucleic Acid Secondary Structure	Compare	and graphically displa and graphically displa	ny two nucleic acid sequences. Ny two peptide sequences.				
Translation Utilities	FrameAlign Creates an op the codons in	timal alignment of the the forward frames of	best segment of similarity (local alignment) between a prot a nucleotide sequence.	ein sequence and			
Index	🎺 <u>Create an</u>	optimal alignment.					
	Gap Uses the algor number match	rithm of Needleman a nes and minimizes the	nd Wunsch to find the alignment of two complete sequences. number of gaps.	It maximizes the			



Globally align two nucleic acid sequences.

Globally align two peptide sequences.







# SeqWeb BestFit Protein Program http://seqweb.stanford.edu:81/gcg-bin/analysis.cgi?program=bestfit-prot

SeqWeb v _{3.1}					Saccelrys
	Programs	Managers			Help Topics   Support
Programs	BestFit				2
Comparison	Locally ali	gn two peptide s	equences.		
Database Searching	Input seque	Select Fi	rom: Default 🛟 Projec	t Local File C	lipboard Database
Similarity	hba hum	uence l nan hba	Description Typ human P	Length 141	Range <u>1141</u>
Reference	Refresh		_numan P	146	<u>1146</u> (Clear)
Evolution					
Mapping	Input Parar	meters:			
Pattern Recognition	Select a se mismatche	quence comparisi s are scored. The each matrix nam	on matrix. This matrix default penalites for e.	determines h gap creation a	ow matches and nd extension are
Primer Selection	Scoring Ma	itrix		blos	um62 🛟
Protein Analysis	Set gap cro	eation penalty		8	
Nucleic Acid	Set gap ex	tension penalty		2	
Secondary	Don't pena	lize gap extension	<u>is longer than</u>		
Structure	Generate s	statistics from 10 r	andomized alignment	<u>s</u> 🛛	
Translation			nucleotide or amino composition	acid 💿	
Utilities	Rando preserving	<u>mize alignment</u> :	dinucleotide or diper	otide 📀	
Index			trinucleotide or tripe composition	ptide O	
	Numbe	er of randomizatio	n <u>s</u>	thru	(range 2
	Run Rese	et		unu	100/

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## BestFit Alignments (Gap =8)

## SeqWeb v_{3.1}

### **BestFit Results**

BESTFIT o	of: <u>hba_human</u>	check:	9231 from: 1 to: 14	1	
WPDEF					
FROMIG of	F:				
	Gap Weight:	8	Average Match:	2.778	
Ler	ngth Weight:	2	Average Mismatch:	-2.248	
	Quality:	286	Length:	145	
	Ratio:	2.058	Gaps:	3	
Percent	Similarity:	51.095	Percent Identity:	45.985	
,	Match display	y thres   = IDE : = 2 . = 1	holds for the alignm NTITY	ent(s):	
<u>hba_huma</u>	an x <u>hbb_hum</u> a	an .	January 31, 2007 21:	17	
2	lspadktnvkad	awgkvgal	hageygaealermflsfptt .          : . :	ktyfphf.dls. . :	49
3	ltpeeksavta	lwgkv	nvdevggealgrllvvypwt	qrffesfgdlst	50
50	hgsaqvkg	ghgkkvad 	daltnavahvddmpnalsal:	sdlhahklrvdp  :	95
51	pdavmgnpkvko	ahgkkvl	gafsdglahldnlkgtfatl:	selhcdklhvdp	100
96	vnfkllshcllv	vtlaahl; 	paeftpavhasldkflasvs        .   .  .	tvltsky 140	
101	enfrllgnvlv	cvlahhf	gkeftppvqaayqkvvagva	nalahky 145	







## BestFit Alignments (Gap =8&2)

Gap Weight: 8 Average Match: 2,778	Gan Weight: 2 Average Match: 2 778
Length Weight: 2 Average Mismatch: -2.248	Length Weight: 1 Average Mismatch: -2.248
Quality: 286 Length: 145	Quality: 313 Length: 147
Ratio: 2.058 Gaps: 3	Ratio: 2.236 Gaps: 4
Percent Similarity: 51.095 Percent Identity: 45.985	Percent Similarity: 51.449 Percent Identity: 46.377
Match display thresholds for the alignment(s):   = IDENTITY	Match display thresholds for the alignment(s):   = IDENTITY
: = 2	: = 2
. = 1	. = 1
hba_human x hbb_human January 31, 2007 21:17	hba_human x hbb_human January 31, 2007 22:31
2 ]spadktpykaawakyaahaaeyaaea]ermf]sfpttktyfphf d]s 49	1 v ] cnodktry (complexes a complexes) or mf] cfort that which f d] 49
	I V.ISpaaktrivkaawgkvganageygaeatermitistpttktytpht.at 46
3 ]theeksavtalwaky nydevagealar]]voormetarffesfad]st 50	1
5 repecksuvearingkv. invacvggeargi revypheqi mesi garse 50	1 vnitpeeksavtaiwgkvnvaevggeaigriivvypwtqrffesfgal 48
50hgsaqvkghgkkvadaltnavahvddmpnalsalsdlhahklrvdp 95	49 shgsaqvkghgkkvadaltnavahvddmpnalsalsdlhahklrv 93
51 pdavmgnpkvkahgkkvlgafsdglahldnlkgtfatlselhcdklhvdp 100	49 stpdavmgnpkvkahgkkvlgafsdglahldnlkgtfatlselhcdklhv 98
96 vnfkllshcllvtlaahlpaeftpavhasldkflasvstvltsky 140	94 dpvnfkllshcllvtlaahlpaeftpavhasldkflasvstvltsky 140
101 enfrllgnvlvcvlahhfgkeftppvqaayqkvvagvanalahky 145	99 dpenfrllgnvlvcvlahhfgkeftppvqaayqkvvagvanalahky 145







## Saccelrys



# SeqWeb Gap Protein Alignments

http://seqweb.stanford.edu:81/gcg-bin/analysis.cgi?program=gap-prot

Gap

#### 1

### Globally align two peptide sequences.

Input sequences: S	Select From: Default 🛟	Project	Local File Clip	board Databa
Sequence	Description	Туре	Length	Range
hba human	hba_human	Р	141	1 141
hbb human	hbb_human	Р	146	1146
Refresh				Cle

#### Input Parameters:

Select a sequence comparision matrix. This matrix determines how matches and mismatches are scored. The default penalites for gap creation and extension are given after each matrix name.

Scoring Matrix		blosum62	+
Set gap creation penalty		8	
Set gap extension penalty		2	
Penalize gans	don't penalize gaps at the ends of the alignment	•	
	penalize end gaps like other gaps	0	
Don't penalize gap extensi	ions longer than		
Generate statistics from 1	0 randomized alignments		
	nucleotide or amino acid composition	•	
Randomize alignment	dinucleotide or dipeptide composition	0	
	trinucleotide or tripeptide composition	0	
Number of randomiza	itions		(range
(Durp (Deret)		thru 100)	
(Kun) (Reset)			



## SeqWeb v_{3.1}



## Gap Results (Gap 8&4)

Gap We	eight: 8	Average Match:	2.778	
Length We	eight: 2	Average Mismatch:	-2.248	
Que	ality: 283	Length:	148	
F	Ratio: 2.007	Gaps:	3	
Percent Simila	arity: 50.360	Percent Identity:	45.324	Pe
Match a	lisplay thres   = IDEN : = 2 . = 1	holds for the alignm NTITY	ent(s):	
<u>hba_human</u> x <u>hb</u>	<u>b_human</u>	January 31, 2007 21:	22	hb
1 .vlspo I.I 1 vhltpe 49 s	adktnvkaawgkvo : .          eeksavtalwgkv hgsaqvkghgkkv	gahageygaealermflsfp .          : . :  nvdevggealgrllvvyp vadaltnavahvddmpnals	ttktyfphf.dl 48 I. :       wtqrffesfgdl 48 alsdlhahklrv 93	
l 49 stpdav	1        /mgnpkvkahgkkv	. .: . vlgafsdglahldnlkgtfa	:        tlselhcdklhv 98	
94 dpvnfk      :	<llshcllvtlaak< td=""><td>hlpaeftpavhasldkflas          .   . </td><td>vstvltskyr 141  .     </td><td></td></llshcllvtlaak<>	hlpaeftpavhasldkflas          .   .	vstvltskyr 141  .	
99 dpenfr	llgnvlvcvlah	hfgkeftppvqaayqkvvag	vanalahkyh 146	

Gan Weight:	4	Average Match:	2.778	
Length Weight:	1	Average Mismatch:	-2.248	
5 5				
Quality:	305	Length:	148	
Ratio:	2.163	Gaps:	4	
rcent Similarity:	51.079	Percent Identity:	46.043	
Match displa	y thresho   = IDEN : = 2 . = 1	olds for the alignm TITY	ent(s):	
a_human x <u>hbb_hum</u>	an Jo	anuary 31, 2007 21:	23	
1 v.lspadktnvl	kaawgkvgo	ahageygaealermflsfp .          : . :	I. :	48
1 vhltpeeksav	talwgkv.	.nvdevggealgrllvvyp	wtqrffesfgdl	48
49 shgsaq I I	vkghgkkva	adaltnavahvddmpnals	alsdlhahklrv   :	93
49 stpdavmgnpk	vkahgkkvi	lgafsdglahldnlkgtfa	tlselhcdklhv	98









# SeqWeb Compare Proteins

http://seqweb.stanford.edu:81/gcg-bin/analysis.cgi?program=compdot-prot

Compare

?

### Compare and graphically display two peptide sequences.

Input sequences: Se	elect From: Default 🛟	Project	Local File Clip	board Database			
Sequence	Description	Туре	Length	Range			
<u>hba human</u>	hba_human	Р	141	<u>1141</u>			
lgba_soybn	lgba_soybn	Р	143	<u>1143</u>			
Refresh							
Input Parameters:			_				
Scoring Matrix			blo	sum30 🛟			
Comparison window			30	•			
Set stringency for mat	ch in comparison win	dow					
Plotting Parameters							
Do not connect adjace	ent points with a line						
Display labels							
Where to Place Tick Numbering bottom Where to Place Tick Numbering right left							
Run Reset							

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## Compare HHA and HHB Human

Comparison Table: share_matrix:blosum62.cmp

BLOSUM62 amino acid substitution matrix. Reference: Henikoff, S. and Henikoff, J. G. (1992). Amino acid substitution matrices from protein blocks. Proc. Natl. Acad. Sci. USA 89: 10915-10919.

Window: 30 Stringency: 10 Points: 151 January 31, 2007 21:24 ...



CC







January 31, 2007 21:26

00TPLOT Density: 184.77

## Compare HHA to Soybean HB

Comparison Table: share_matrix:blosum30.cmp

BLOSUM30 amino acid substitution matrix.

Window: 30 Stringency: 15 Points: 469 January 31, 2007 21:26 ...



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