Gathering Sequences: BLAST

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Searching................................................done
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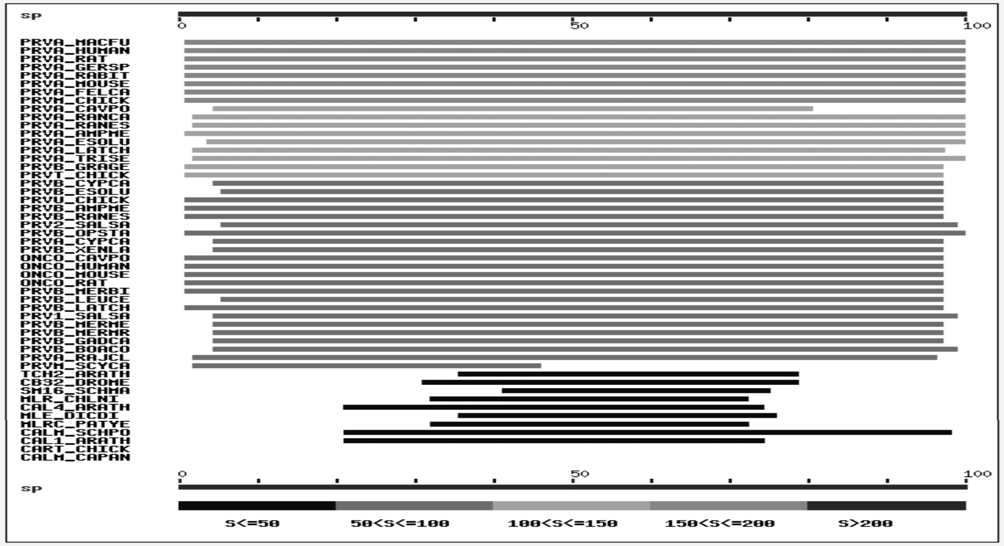


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                                                                 Score    E
Sequences producing significant alignments:                      (bits) Value

sp|P80050|PRVA_MACFU  (PVALB)PARVALBUMIN ALPHA (PARVALBUMIN, MUS...  187  7e-48
sp|P20472|PRVA_HUMAN  (PVALB)PARVALBUMIN ALPHA.[Homo sapiens]        181  3e-46
sp|P02625|PRVA_RAT  (PVALB..)PARVALBUMIN ALPHA.[Rattus norvegicus]   175  2e-44
sp|P80080|PRVA_GERSP  PARVALBUMIN ALPHA.[Gerbillus sp.]              173  1e-43
sp|P02624|PRVA_RABIT  (PVALB)PARVALBUMIN ALPHA.[Oryctolagus cuni...  172  2e-43
sp|P32848|PRVA_MOUSE  (PVALB..)PARVALBUMIN ALPHA.[Mus musculus]      168  3e-42
sp|P80079|PRVA_FELCA  (PVALB)PARVALBUMIN ALPHA.[Felis silvestris...  163  8e-41
sp|P80026|PRVM_CHICK  PARVALBUMIN, MUSCLE.[Gallus gallus]            154  5e-38
sp|P51434|PRVA_CAVPO  (PVALB)PARVALBUMIN ALPHA (FRAGMENT).[Cavia...  132  1e-31
sp|P18087|PRVA_RANCA  PARVALBUMIN ALPHA (PA 4.97).[Rana catesbei...  123  1e-28
sp|P02627|PRVA_RANES  PARVALBUMIN ALPHA.[Rana esculenta]             121  4e-28
sp|P02626|PRVA_AMPME  PARVALBUMIN ALPHA.[Amphiuma means]             111  4e-25
sp|P02628|PRVA_ESOLU  PARVALBUMIN ALPHA.[Esox lucius]                111  5e-25
sp|P02614|PRVB_GRAGE  PARVALBUMIN BETA.[Graptemys geographica]       107  6e-24
sp|P30563|PRVA_TRISE  PARVALBUMIN ALPHA.[Triakis semifasciata]       107  6e-24
sp|P02544|MLRC_PATYE  MYOSIN REGULATORY LIGHT CHAIN, STRIATED AD...   35  0.038
sp|P05963|MLR_CHLNI  MYOSIN REGULATORY LIGHT CHAIN, STRIATED ADD...   36  0.022
sp|Q07167|SM16_SCHMA  16 KDA CALCIUM-BINDING PROTEIN (EGG ANTIGE...   37  0.010
sp|P41044|CB32_DROME  (CBP53E..)CALBINDIN-32.[Drosophila melanog...   39  0.003
sp|P25070|TCH2_ARATH  (TCH2)CALMODULIN-RELATED PROTEIN 2, TOUCH-...   40  0.002
sp|P32461|PRVM_SCYCA  PARVALBUMIN (FRAGMENT).[Scyliorhinus canic...   68  7e-12
sp|P02630|PRVA_RAJCL  PARVALBUMIN ALPHA.[Raja clavata]                71  6e-13
sp|P02615|PRVB_BOACO  PARVALBUMIN BETA.[Boa constrictor]              75  4e-14
sp|P02622|PRVB_GADCA  PARVALBUMIN BETA (ALLERGEN GAD C 1) (GAD C...   75  3e-14
sp|P02621|PRVB_MERMR  PARVALBUMIN BETA.[Merlangius merlangus]         80  1e-15
sp|P02620|PRVB_MERME  PARVALBUMIN BETA.[Merluccius merluccius]        83  1e-16
sp|Q91482|PRV1_SALSA  PARVALBUMIN BETA 1 (CLONE 14.1) (MAJOR ALL...   86  2e-17
sp|P02623|PRVB_LATCH  PARVALBUMIN BETA.[Latimeria chalumnae]          87  1e-17
sp|P02631|ONCO_RAT  (OCM)ONCOMODULIN (OM) (PARVALBUMIN BETA).[Ra...   88  4e-18
sp|P05939|PRVB_LEUCE  PARVALBUMIN BETA (A).[Leuciscus cephalus]       88  4e-18
sp|P56503|PRVB_MERBI  PARVALBUMIN BETA.[Merluccius bilinearis]        88  4e-18
sp|P51879|ONCO_MOUSE  (OCM)ONCOMODULIN (OM) (PARVALBUMIN BETA).[...   89  3e-18
sp|P05940|PRVB_XENLA  PARVALBUMIN BETA.[Xenopus laevis]               89  3e-18
sp|O35508|ONCO_CAVPO  (OCM)ONCOMODULIN (OM) (PARVALBUMIN BETA) (...   89  3e-18
sp|P32930|ONCO_HUMAN  (OCM..)ONCOMODULIN (OM) (PARVALBUMIN BETA)...   89  2e-18
sp|P09227|PRVA_CYPCA  PARVALBUMIN ALPHA (A1).[Cyprinus carpio]        90  1e-18
sp|P05941|PRVB_OPSTA  PARVALBUMIN BETA (PARVALBUMIN III).[Opsan...    93  1e-19
sp|Q91483|PRV2_SALSA  PARVALBUMIN BETA 2 (CLONE 24.1) (MAJOR ALL...   94  8e-20
sp|P02617|PRVB_RANES  PARVALBUMIN BETA.[Rana esculenta]               94  8e-20
sp|P02616|PRVB_AMPME  PARVALBUMIN BETA.[Amphiuma means]               94  6e-20
sp|P43305|PRVU_CHICK  PARVALBUMIN, THYMIC CPV3 (PARVALBUMIN 3).[...   95  3e-20
sp|P02619|PRVB_ESOLU  PARVALBUMIN BETA.[Esox lucius]                  97  1e-20
sp|P02618|PRVB_CYPCA  PARVALBUMIN BETA.[Cyprinus carpio]              99  2e-21
```

Common Mistake: Sequences Too Closely Related

```
PRVA_MACFU   SMTDLLNAEDIKKAVGAFSAIDSFDHKKFFQMVGLKKKSADDVKKVFHILDKDKSGFIEE
PRVA_HUMAN   SMTDLLNAEDIKKAVGAFSATDSFDHKKFFQMVGLKKKSADDVKKVFHMLDKDKSGFIEE
PRVA_GERSP   SMTDLLSAEDIKKAIGAFAAADSFDHKKFFQMVGLKKKTPDDVKKVFHILDKDKSGFIEE
PRVA_MOUSE   SMTDVLSAEDIKKAIGAFAAADSFDHKKFFQMVGLKKKNPDEVKKVFHILDKDKSGFIEE
PRVA_RAT     SMTDLLSAEDIKKAIGAFTAADSFDHKKFFQMVGLKKKSADDVKKVFHILDKDKSGFIEE
PRVA_RABIT   AMTELLNAEDIKKAIGAFAAAESFDHKKFFQMVGLKKKSTEDVKKVFHILDKDKSGFIEE
              :**::*.*******:***:*  :*****************..::******:***********

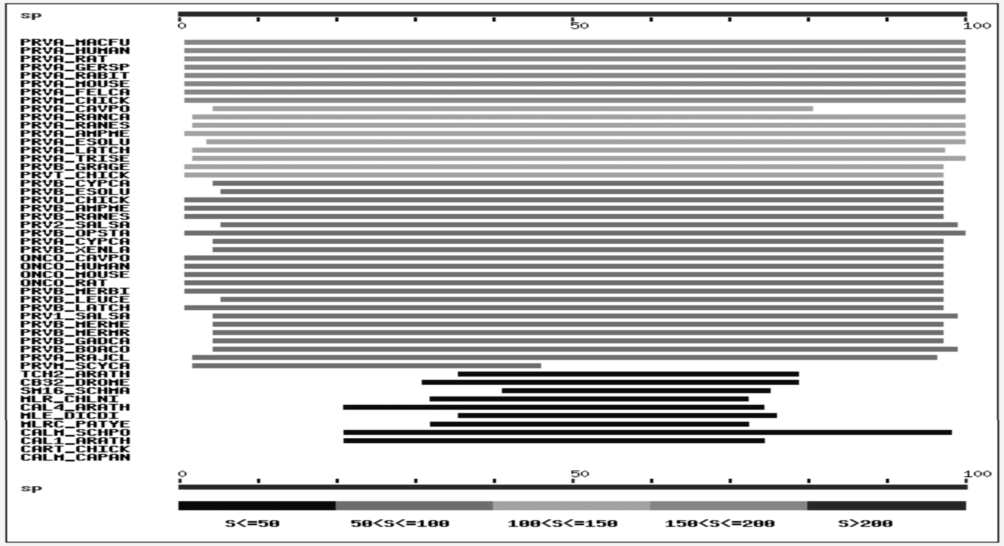
PRVA_MACFU   DELGFILKGFSPDARDLSAKETKTLMAAGDKDGDGKIGVDEFSTLVAES
PRVA_HUMAN   DELGFILKGFSPDARDLSAKETKMLMAAGDKDGDGKIGVDEFSTLVAES
PRVA_GERSP   DELGFILKGFSSDARDLSAKETKTLLAAGDKDGDGKIGVEEFSTLVSES
PRVA_MOUSE   DELGSILKGFSSDARDLSAKETKTLLAAGDKDGDGKIGVEEFSTLVAES
PRVA_RAT     DELGSILKGFSSDARDLSAKETKTLMAAGDKDGDGKIGVEEFSTLVAES
PRVA_RABIT   EELGFILKGFSPDARDLSVKETKTLMAAGDKDGDGKIGADEFSTLVSES
              :*** ******.******.**** *.************.:******:**
```

-IDENTICAL SEQUENCES BRING NO INFORMATION FOR THE MULTIPLE SEQUENCE ALIGNMENT

-MULTIPLE SEQUENCE ALIGNMENTS THRIVE ON DIVERSITY…

Selecting Diverse Sequences (Opus I)

Searching..done



Sequences producing significant alignments:	Score (bits)	E Value
sp|P80050|PRVA MACFU (PVALB)PARVALBUMIN ALPHA (PARVALBUMIN, MUS...	187	7e-48
sp|P20472|PRVA HUMAN (PVALB)PARVALBUMIN ALPHA.[Homo sapiens]	181	3e-46
sp|P02625|PRVA RAT (PVALB..)PARVALBUMIN ALPHA.[Rattus norvegicus]	175	2e-44
sp|P80080|PRVA GERSP PARVALBUMIN ALPHA.[Gerbillus sp.]	173	1e-43
sp|P02624|PRVA RABIT (PVALB)PARVALBUMIN ALPHA.[Oryctolagus cuni...	172	2e-43
sp|P32848|PRVA MOUSE (PVALB..)PARVALBUMIN ALPHA.[Mus musculus]	168	3e-42
sp|P80079|PRVA FELCA (PVALB)PARVALBUMIN ALPHA.[Felis silvestris...	163	8e-41
sp|P80026|PRVM CHICK PARVALBUMIN, MUSCLE.[Gallus gallus]	154	5e-38
sp|P51434|PRVA CAVPO (PVALB)PARVALBUMIN ALPHA (FRAGMENT).[Cavia...	132	1e-31
sp|P18087|PRVA RANCA PARVALBUMIN ALPHA (PA 4.97).[Rana catesbei...	123	1e-28
sp|P02627|PRVA RANES PARVALBUMIN ALPHA.[Rana esculenta]	121	4e-28
sp|P02626|PRVA AMPME PARVALBUMIN ALPHA.[Amphiuma means]	111	4e-25
sp|P02628|PRVA ESOLU PARVALBUMIN ALPHA.[Esox lucius]	111	5e-25
sp|P02614|PRVB GRAGE PARVALBUMIN BETA.[Graptemys geographica]	107	6e-24
sp|P30563|PRVA TRISE PARVALBUMIN ALPHA.[Triakis semifasciata]	107	6e-24
sp|P05944|MLRC PATYE MYOSIN REGULATORY LIGHT CHAIN, STRIATED AD...	35	0.038
sp|P05963|MLR CHLNI MYOSIN REGULATORY LIGHT CHAIN, STRIATED ADD...	36	0.022
sp|Q02157|SM16 SCHMA 16 KDA CALCIUM-BINDING PROTEIN (EGG ANTIGE...	37	0.010
sp|P41044|CB32 DROME (CBP53E..)CALBINDIN-32.[Drosophila melanog...	39	0.003
sp|P25070|TCH2 ARATH (TCH2)CALMODULIN-RELATED PROTEIN 2, TOUCH-...	40	0.002
sp|P32481|PRVM SCYCA PARVALBUMIN (FRAGMENT).[Scyliorhinus canic...	68	7e-12
sp|P02630|PRVA RAJCL PARVALBUMIN ALPHA.[Raja clavata]	71	6e-13
sp|P02615|PRVB BOACO PARVALBUMIN BETA.[Boa constrictor]	75	4e-14
sp|P02622|PRVB GADCA PARVALBUMIN BETA (ALLERGEN GAD C 1) (GAD C...	75	3e-14
sp|P02621|PRVB MERMR PARVALBUMIN BETA.[Merlangius merlangus]	80	1e-15
sp|P02620|PRVB MERME PARVALBUMIN BETA.[Merluccius merluccius]	83	1e-16
sp|Q91482|PRV1 SALSA PARVALBUMIN BETA 1 (CLONE 14.1) (MAJOR ALL...	86	2e-17
sp|P02623|PRVB LATCH PARVALBUMIN BETA.[Latimeria chalumnae]	87	1e-17
sp|P02631|ONCO RAT (OCM)ONCOMODULIN (OM) (PARVALBUMIN BETA).[Ra...	88	4e-18
sp|P02639|PRVB LEUCE PARVALBUMIN BETA (V).[Leuciscus cephalus]	88	4e-18
sp|P56503|PRVB MERBI PARVALBUMIN BETA.[Merluccius bilinearis]	88	4e-18
sp|P51879|ONCO MOUSE (OCM)ONCOMODULIN (OM) (PARVALBUMIN BETA).[...	89	3e-18
sp|P02640|PRVB XENLA PARVALBUMIN BETA.[Xenopus laevis]	89	3e-18
sp|O35508|ONCO CAVPO (OCM)ONCOMODULIN (OM) (PARVALBUMIN BETA) (...	89	3e-18
sp|P32930|ONCO HUMAN (OCM..)ONCOMODULIN (OM) (PARVALBUMIN BETA)...	89	2e-18
sp|P09227|PRVA CYPCA PARVALBUMIN ALPHA (A1).[Cyprinus carpio]	90	1e-18
sp|P02641|PRVB OPSTA PARVALBUMIN BETA (PARVALBUMIN IIIB).[Opsan...	93	1e-19
sp|Q91483|PRV2 SALSA PARVALBUMIN BETA 2 (CLONE 24.1) (MAJOR ALL...	94	8e-20
sp|P02617|PRVB RANES PARVALBUMIN BETA.[Rana esculenta]	94	8e-20
sp|[illegible]|PRVB AMPME PARVALBUMIN BETA.[Amphiuma means]	[illegible]	[illegible]
sp|P43305|PRVU CHICK PARVALBUMIN, THYMIC CPV3 (PARVALBUMIN 3).[...	95	3e-20
sp|P02619|PRVB ESOLU PARVALBUMIN BETA.[Esox lucius]	97	1e-20
sp|P02618|PRVB CYPCA PARVALBUMIN BETA.[Cyprinus carpio]	99	2e-21

Respect Information!

```
PRVA_MACFU   ------------------------------------------SMTDLLN----AEDIKKA
PRVA_HUMAN   ------------------------------------------SMTDLLN----AEDIKKA
PRVA_GERSP   ------------------------------------------SMTDLLS----AEDIKKA
PRVA_MOUSE   ------------------------------------------SMTDVLS----AEDIKKA
PRVA_RAT     ------------------------------------------SMTDLLS----AEDIKKA
PRVA_RABIT   ------------------------------------------AMTELLN----AEDIKKA
TPCC_MOUSE   MDDIYKAAVEQLTEEQKNEFKAAFDIFVLGAEDGCISTKELGKVMRMLGQNPTPEELQEM
                                                       :  :*.     .*::::

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PRVA_HUMAN   VGAFSATDS--FDHKKFFQMVG------LKKKSADDVKKVFHMLDKDKSGFIEEDELGFI
PRVA_GERSP   IGAFAAADS--FDHKKFFQMVG------LKKKTPDDVKKVFHILDKDKSGFIEEDELGFI
PRVA_MOUSE   IGAFAAADS--FDHKKFFQMVG------LKKKNPDEVKKVFHILDKDKSGFIEEDELGSI
PRVA_RAT     IGAFTAADS--FDHKKFFQMVG------LKKKSADDVKKVFHILDKDKSGFIEEDELGSI
PRVA_RABIT   IGAFAAAES--FDHKKFFQMVG------LKKKSTEDVKKVFHILDKDKSGFIEEEELGFI
TPCC_MOUSE   IDEVDEDGSGTVDFDEFLVMMVRCMKDDSKGKSEEELSDLFRMFDKNADGYIDLDELKMM
             :.  .     *  .*..:*: *:        * *. ::::..:*:::**: .*:*: :**  :

PRVA_MACFU   LKGFSPDARDLSAKETKTLMAAGDKDGDGKIGVDEFSTLVAES-
PRVA_HUMAN   LKGFSPDARDLSAKETKMLMAAGDKDGDGKIGVDEFSTLVAES-
PRVA_GERSP   LKGFSSDARDLSAKETKTLLAAGDKDGDGKIGVEEFSTLVSES-
PRVA_MOUSE   LKGFSSDARDLSAKETKTLLAAGDKDGDGKIGVEEFSTLVAES-
PRVA_RAT     LKGFSSDARDLSAKETKTLMAAGDKDGDGKIGVEEFSTLVAES-
PRVA_RABIT   LKGFSPDARDLSVKETKTLMAAGDKDGDGKIGADEFSTLVSES-
TPCC_MOUSE   LQ---ATGETITEDDIEELMKDGDKNNDGRIDYDEFLEFMKGVE
             *:    . .. :: .: : *:  ***:.**:*. :**   ::
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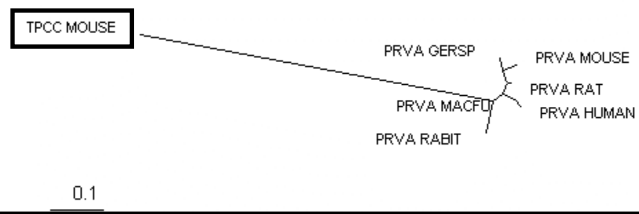
-This Alignment Is not Informative about the relation Betwwen TPCC MOUSE and the rest of the sequences.

-A better Spread of the Sequences is needed



Selecting Diverse Sequences (Opus II)

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Searching.................................................done
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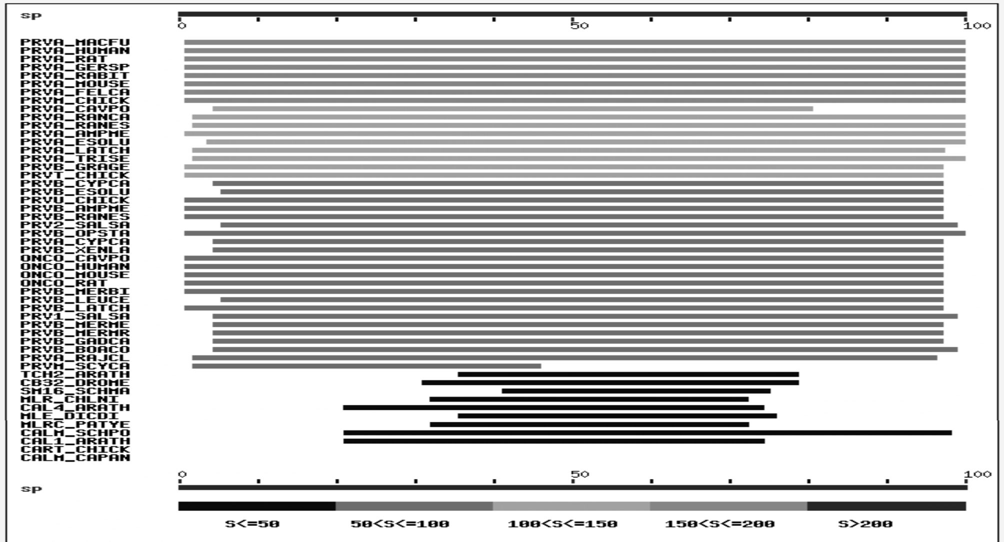

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                                                                 Score     E
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sp|P30563|PRVA TRISE  PARVALBUMIN ALPHA.[Triakis semifasciata]      107   6e-24
sp|Q07167|SM16 SCHMA  16 KDA CALCIUM-BINDING PROTEIN (EGG ANTIGE...  37   0.010
sp|P41044|CB32 DROME  (CBP32E..)CALBINDIN-32.[Drosophila melanog...  39   0.003
sp|P25070|TCH2 ARATH  (TCH2)CALMODULIN-RELATED PROTEIN 2, TOUCH-...  40   0.002
sp|P32461|PRVM SCYCA  PARVALBUMIN (FRAGMENT).[Scyliorhinus canic...  68   7e-12
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sp|P02620|PRVB MERME  PARVALBUMIN BETA.[Merluccius merluccius]       83   1e-16
sp|Q91482|PRV1 SALSA  PARVALBUMIN BETA 1 (CLONE 14.1) (MAJOR ALL...  86   2e-17
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sp|P02641|PRVB OPSTA  PARVALBUMIN BETA (PARVALBUMIN IIIB).[Opsan...  93   1e-19
sp|Q91483|PRV2 SALSA  PARVALBUMIN BETA 2 (CLONE 24.1) (MAJOR ALL...  94   8e-20
sp|P02617|PRVB RANES  PARVALBUMIN BETA.[Rana esculenta]              94   8e-20
sp|P02616|PRVB AMPME  PARVALBUMIN BETA.[Amphiuma means]              94   9e-20
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sp|P02619|PRVB ESOLU  PARVALBUMIN BETA.[Esox lucius]                 97   1e-20
sp|P02618|PRVB CYPCA  PARVALBUMIN BETA.[Cyprinus carpio]             99   2e-21
```

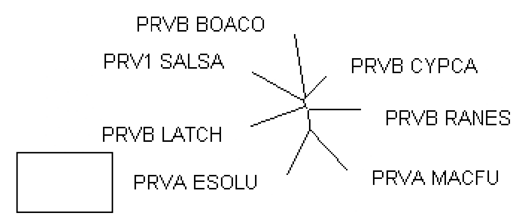
Selecting Diverse Sequences (Opus II)

```
PRVB_CYPCA   -AFAGVLNDADIAAALEACKAADSFNHKAFFAKVGLTSKSADDVKKAFAIIDQDKSGFIE
PRVB_BOACO    -AFAGILSDADIAAGLQSCQAADSFSCKTFFAKSGLHSKSKDQLTKVFGVIDRDKSGYIE
PRV1_SALSA    MACAHLCKEADIKTALEACKAADTFSFKTFFHTIGFASKSADDVKKAFKVIDQDASGFIE
PRVB_LATCH    -AVAKLLAAADVTAALEGCKADDSFNHKVFFQKTGLAKKSNEELEAIFKILDQDKSGFIE
PRVB_RANES    -SITDIVSEKDIDAALESVKAAGSFNYKIFFQKVGLAGKSAADAKKVFEILDRDKSGFIE
PRVA_MACFU    -SMTDLLNAEDIKKAVGAFSAIDSFDHKKFFQMVGLKKKSADDVKKVFHILDKDKSGFIE
PRVA_ESOLU    --AKDLLKADDIKKALDAVKAEGSFNHKKFFALVGLKAMSANDVKKVFKAIDADASGFIE
                    :   *:  :  :  *  :*  * **   *:   *  :   *  :* * **:**

PRVB_CYPCA   EDELKLFLQNFKADARALTDGETKTFLKAGDSDGDGKIGVDEFTALVKA-
PRVB_BOACO    EDELKKFLQNFDGKARDLTDKETAEFLKEGDTDGDGKIGVEEFVVLVTKG
PRV1_SALSA    VEELKLFLQNFCPKARELTDAETKAFLKAGDADGDGMIGIDEFAVLVKQ-
PRVB_LATCH    DEELELFLQNFSAGARTLTKTETETFLKAGDSDGDGKIGVDEFQKLVKA-
PRVB_RANES    QDELGLFLQNFRASARVLSDAETSAFLKAGDSDGDGKIGVEEFQALVKA-
PRVA_MACFU    EDELGFILKGFSPDARDLSAKETKTLMAAGDKDGDGKIGVDEFSTLVAES
PRVA_ESOLU    EEELKFVLKSFAADGRDLTDAETKAFLKAADKDGDGKIGIDEFETLVHEA
               :**  .*:.*   .* *:  **  ::  .* **** **::**  **
```

-A REASONABLE Model Now Exists.

-Going Further:Remote Homologues.



Aligning Remote Homologues

```
PRVA_MACFU   -----------------------------------------SMTDLLNA----EDIKKA
PRVA_ESOLU   ------------------------------------------AKDLLKA----DDIKKA
PRVB_CYPCA   -----------------------------------------AFAGVLND----ADIAAA
PRVB_BOACO   -----------------------------------------AFAGILSD----ADIAAG
PRV1_SALSA   ----------------------------------------MACAHLCKE----ADIKTA
PRVB_LATCH   -----------------------------------------AVAKLLAA----ADVTAA
PRVB_RANES   -----------------------------------------SITDIVSE----KDIDAA
TPCS_RABIT   -TDQQAEARSYLSEEMIAEFKAAFDMFDADGG-GDISVKELGTVMRMLGQTPTKEELDAI
TPCS_PIG     -TDQQAEARSYLSEEMIAEFKAAFDMFDADGG-GDISVKELGTVMRMLGQTPTKEELDAI
TPCC_MOUSE   MDDIYKAAVEQLTEEQKNEFKAAFDIFVLGAEDGCISTKELGKVMRMLGQNPTPEELQEM
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PRVA_MACFU   VGAFSAIDS--FDHKKFFQMVG------LKKKSADDVKKVFHILDKDKSGFIEEDELGFI
PRVA_ESOLU   LDAVKAEGS--FNHKKFFALVG------LKAMSANDVKKVFKAIDADASGFIEEEELKFV
PRVB_CYPCA   LEACKAADS--FNHKAFFAKVG------LTSKSADDVKKAFAIIDQDKSGFIEEDELKLF
PRVB_BOACO   LQSCQAADS--FSCKTFFAKSG------LHSKSKDQLTKVFGVIDRDKSGYIEEDELKKF
PRV1_SALSA   LEACKAADT--FSFKTFFHTIG------FASKSADDVKKAFKVIDQDASGFIEVEELKLF
PRVB_LATCH   LEGCKADDS--FNHKVFFQKTG------LAKKSNEELEAIFKILDQDKSGFIEDEELELF
PRVB_RANES   LESVKAAGS--FNYKIFFQKVG------LAGKSAADAKKVFEILDRDKSGFIEQDELGLF
TPCS_RABIT   IEEVDEDGSGTIDFEEFLVMMVRQMKEDAKGKSEEELAECFRIFDRNADGYIDAEELAEI
TPCS_PIG     IEEVDEDGSGTIDFEEFLVMMVRQMKEDAKGKSEEELAECFRIFDRNMDGYIDAEELAEI
TPCC_MOUSE   IDEVDEDGSGTVDFDEFLVMMVRCMKDDSKGKSEEELSDLFRMFDKNADGYIDLDELKMM
             :   .  .:  . . . *:                  *  :     *  :* : .*:*: :**  .

PRVA_MACFU      LKGFSPDARDLSAKETKTLMAAGDKDGDGKIGVDEFSTLVAES-
PRVA_ESOLU      LKSFAADGRDLTDAETKAFLKAADKDGDGKIGIDEFETLVHEA-
PRVB_CYPCA      LQNFKADARALTDGETKTFLKAGDSDGDGKIGVDEFTALVKA--
PRVB_BOACO      LQNFDGKARDLTDKETAEFLKEGDTDGDGKIGVEEFVVLVTKG-
PRV1_SALSA      LQNFCPKARELTDAETKAFLKAGDADGDGMIGIDEFAVLVKQ--
PRVB_LATCH      LQNFSAGARTLTKTETETFLKAGDSDGDGKIGVDEFQKLVKA--
PRVB_RANES      LQNFRASARVLSDAETSAFLKAGDSDGDGKIGVEEFQALVKA--
TPCS_RABIT      FR---ASGEHVTDEEIESLMKDGDKNNDGRIDFDEFLKMMEGVQ
TPCS_PIG        FR---ASGEHVTDEEIESIMKDGDKNNDGRIDFDEFLKMMEGVQ
TPCC_MOUSE      LQ---ATGETITEDDIEELMKDGDKNNDGRIDYDEFLEFMKGVE
                ::        .. ::  :    :: .* : .** *.  :**    ::
```

Going Further…

General information about the entry	
Entry name	TPC_PATYE
Primary accession number	P35622

Comments

- *FUNCTION*: TROPONIN IS THE CENTRAL REGULATORY PROTEIN OF STRIATED MUSCLE CONTRACTION. TN CONSISTS OF THREE COMPONENTS: TN-I WHICH IS THE INHIBITOR OF ACTOMYOSIN ATPASE, TN-T WHICH CONTAINS THE BINDING SITE FOR TROPOMYOSIN AND TN-C. THE BINDING OF CALCIUM TO TN-C ABOLISHES THE INHIBITORY ACTION OF TN ON ACTIN FILAMENTS.
- *MISCELLANEOUS*: THIS PROTEIN BINDS ONE CALCIUM ION PER MOLECULE.
- *SIMILARITY*: TO OTHER EF-HAND CALCIUM BINDING PROTEINS.

Features

MOD_RES	1	1	ACETYLATION.
DOMAIN	22	33	ANCESTRAL CALCIUM SITE 1.
DOMAIN	58	69	ANCESTRAL CALCIUM SITE 2.
DOMAIN	95	106	ANCESTRAL CALCIUM SITE 3.
CA_BIND	131	142	SITE 4.

```
PRVA_MACFU   VGAFSAIDS--FDHKKFFQMVG------LKKKSADDVKKVFHILDKDKSGFIEEDELGFI
PRVB_BOACO   LQSCQAADS--FSCKTFFAKSG------LHSKSKDQLTKVFGVIDRDKSGYIEEDELKKF
PRV1_SALSA   LEACKAADT--FSFKTFFHTIG------FASKSADDVKKAFKVIDQDASGFIEVEELKLF
TPCS_RABIT   IEEVDEDGSGTIDFEEFLVMMVRQMKEDAKGKSEEELAECFRIFDRNADGYIDAEELAEI
TPCS_PIG     IEEVDEDGSGTIDFEEFLVMMVRQMKEDAKGKSEEELAECFRIFDRNMDGYIDAEELAEI
TPCC_MOUSE   IDEVDEDGSGTVDFDEFLVMMVRCMKDDSKGKSEEELSDLFRMFDKNADGYIDLDELKMM
TPC_PATYE    SDEMDEEATGRLNCDAWIQLFER---KLKEDLDERELKEAFRVLDKEKKGVIKVDVLRWI
                  .    :  .. .  ::             .   :    *   :* :  .* *. : *  .

PRVA_MACFU   LKGFSPDARDLSAKETKTLMAAGDKDGDGKIGVDEFSTLVAES--
PRVB_BOACO   LQNFDGKARDLTDKETAEFLKEGDTDGDGKIGVEEFVVLVTKG--
PRV1_SALSA   LQNFCPKARELTDAETKAFLKAGDADGDGMIGIDEFAVLVKQ---
TPCS_RABIT   FR---ASGEHVTDEEIESLMKDGDKNNDGRIDFDEFLKMMEGVQ-
TPCS_PIG     FR---ASGEHVTDEEIESIMKDGDKNNDGRIDFDEFLKMMEGVQ-
TPCC_MOUSE   LQ---ATGETITEDDIEELMKDGDKNNDGRIDYDEFLEFMKGVE-
TPC_PATYE    LS---SLGDELTEEEIENMIAETDTDGSGTVDYEEFKCLMMSSDA
             :       .  ::  :    ::   * :..* :.  :**   ::
```

WHAT MAKES A GOOD ALIGNMENT…

-THE MORE DIVERGEANT THE SEQUENCES, THE BETTER

-THE FEWER INDELS, THE BETTER

-NICE UNGAPPED BLOCKS SEPARATED WITH INDELS

-DIFFERENT CLASSES OF RESIDUES WITHIN A BLOCK:

- Completely Conserved
- Conserved For Size *and* Hydropathy
- Conserved For Size *or* Hydropathy

-THE ULTIMATE EVALUATION IS A MATTER OF PERSONNAL JUDGEMENT AND KNOWLEDGE.

DO NOT OVERTUNE!!!

```
chite   ---ADKPKRPLSAYMLWLNSARESIKRENPDFK-VTEVAKKGGELWRGLKD
wheat   --DPNKPKRAPSAFFVFMGEFREEFKQKNPKNKSVAAVGKAAGERWKSLSE
trybr   KKDSNAPKRAMTSFMFFSSDFRS----KHSDLS-IVEMSKAAGAAWKELGP
mouse   -----KPKRPRSAYNIYVSESFQ----EAKDDS-AQGKLKLVNEAWKNLSP
              ***. ::: .: ..  .    :  . .      *  .  *: *

chite   AATAKQNYIRALQEYERNGG-
wheat   ANKLKGEYNKAIAAYNKGESA
trybr   AEKDKERYKREM---------
mouse   AKDDRIRYDNEMKSWEEQMAE
        *   : .* . :
```

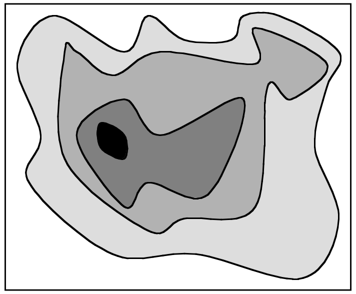
DO NOT PLAY WITH PARAMETERS IF YOU KNOW THE ALIGNMENT YOU WANT: MAKE IT YOURSELF!

```
chite   ---ADKPKRPL-SAYMLWLNSARESIKRENPDFK-VTEVAKKGGELWRGLKD
wheat   --DPNKPKRAP-SAFFVFMGEFREEFKQKNPKNKSVAAVGKAAGERWKSLSE
trybr   KKDSNAPKRAMTSFMFFSSDFRS----KHSDLS-IVEMSKAAGAAWKELGP
mouse   -----KPKRPR-SAYNIYVSESFQ----EAKDDS-AQGKLKLVNEAWKNLSP
              ***. :*: .: ..  .    :  . .      *  .  *: *

chite   AATAKQNYIRALQEYERNGG-
wheat   ANKLKGEYNKAIAAYNKGESA
trybr   AEKDKERYKREM---------
mouse   AKDDRIRYDNEMKSWEEQMAE
        *   : .* . :
```

TUNING or NOT TUNING?

-PARAMETERS TO TUNE USUALLY INCLUDE:
- GOP/ GEP
- MATRIX
- SENSITIVITY Vs SPEED



Substitution Matrices (Etzold and al. 1993)

Gonnet	61.7
Blosum50	59.7
Pam250	59.2

-MOST METHODS ARE TUNED FOR WORKING WELL ON AVERAGE

-PARAMETERS BEHAVIOUR DO NOT NECESSARILY FOLLOW THE THEORY (i.e. Substitution Matrices).

-A GOOD ALIGNMENT IS USUALLY ROBUST(i.e. Changes little).

-TUNE IF YOU WANT TO CONVINCE YOURSELF.

KEEP A BIOLOGICAL PERSPECTIVE

```
chite  ---ADKPKRPLSAYMLWLNSARESIKRENPDFK-VTEVAKKGGELWRGLKD
wheat  --DPNKPKRAPSAFFVFMGEFREEFKQKNPKNKSVAAVGKAAGERWKSLSE
trybr  KKDSNAPKRAMTSFMFFSSDFRS----KHSDLS-IVEMSKAAGAAWKELGP
mouse  -----KPKRPRSAYNIYVSESFQ----EAKDDS-AQGKLKLVNEAWKNLSP
            ***. ::: .: ..  .    :  .  .      *  .  *: *

chite  AATAKQNYIRALQEYERNGG-
wheat  ANKLKGEYNKAIAAYNKGESA
trybr  AEKDKERYKREM---------
mouse  AKDDRIRYDNEMKSWEEQMAE
       *   : .* . :
```

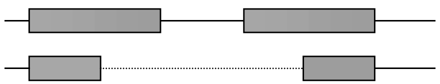
DIFFERENT PARAMETERS

```
chite AD--K----PKR-PLYMLWLNS-ARESIKRENPDFK-VT-EVAKKGGELWRGL-
wheat -DPNK----PKRAP-FFVFMGE-FREEFKQKNPKNKSVA-AVGKAAGERWKSLS
trybr -K--KDSNAPKR-AMT-MFFSSDFR-S-KH-S-DLS-IV-EMSKAAGAAWKELG
mouse ----K----PKR-PRYNIYVSESFQEA-K--D-D-S-AQGKL-KLVNEAWKNLS
          *    *** .:: ::...  :   *  . . .     : *  .  *: *

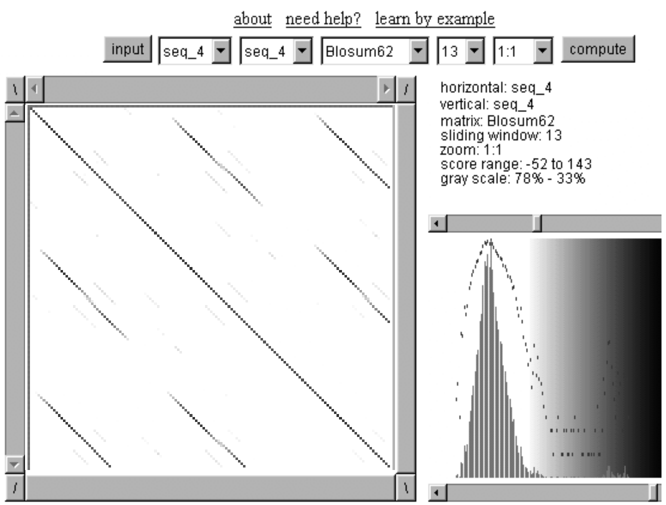
chite KSEWEAKAATAKQNY-I--RALQE-YERNG-G-
wheat KAPYVAKANKLKGEY-N--KAIAA-YNK-GESA
trybr RKVYEEMAEKDKERY----K--RE-M-------
mouse KQAYIQLAKDDRIRYDNEMKSWEEQMAE-----
      :  :   *   : .*     :
```

REPEATS

THERE IS A PROBLEM WHEN TWO SEQUENCES DO NOT CONTAIN THE SAME NUMBER OF REPEATS

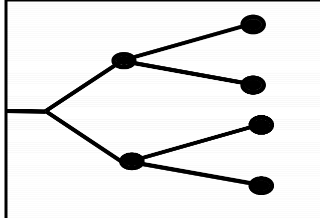
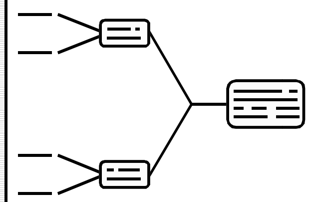
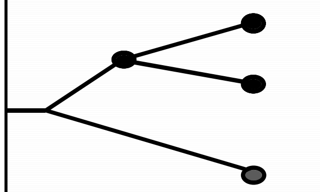
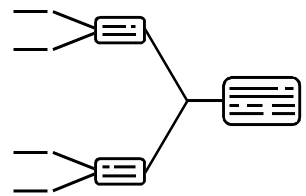
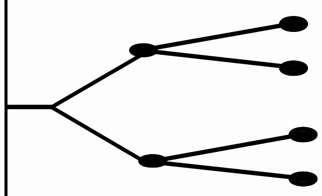
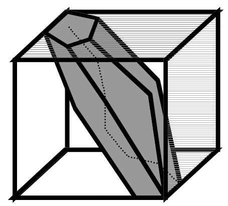
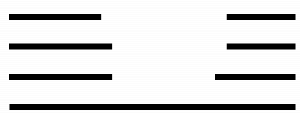
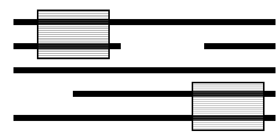
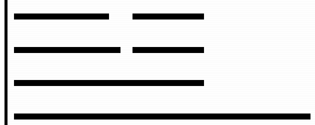
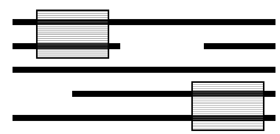


IT IS THEN BETTER TO MANUALLY EXTRACT THE REPEATS AND TO ALIGN THEM. INDIVIDUAL REPEATS CAN BE RECOGNIZED USING DOTTER



Choosing The Right Method

Source: BaliBase, Thompson et al, NAR, 1999

PROBLEM	PROGRAM	METHOD
	ClustalW	
	ClustalW	
	MSA	
	DIALIGN II	
	DIALIGN II	

Examples of Mistakes

Playing With Blocks: Mbh1

```
              Basic - Helix I          Loop        Helix II
primer        -----
Mbh1     SNILERNKARDLALAIRDSER          3    AQVEIITDGEEPAEMIQVLGPK
c-Myc    HNVLERQRRNELKRSFFALRD         11    PKVVILKKATAYILSIQADEHK
TFE-3    HNLIERRRRFNINDRIKELGT         12    NKGTILKASVDYIRKLQKEQQR
E12      NNARERLRVRDINEAFKELGR         12    TKLLILHQAVSVILNLEQQVRE
MyoD     ATMRERRRLSKVNEAFETLKR         10    PKVEILRNAIRYIEGLQALLRD
         "maybe"                             5.4e-3
         "maybe"                     4-12    1
         "no"
         "no"
```

Figure 2. Evaluation of block alignments involving Mbh1 using the MACAW program (Schuler et al., 1991) as described by Prendergast and Ziff (1991).

Blocks are reproduced as in their Figure 1 except that three conflicting residues in the second block for c-Myc are taken from the reference that they cite. MACAW scores a block as "yes," "no," or "maybe" depending on how the block compares to a random protein model. Brackets show the interval and MACAW evaluation for the first block, where successive columns of primer-related sequences are trimmed off. For the second block, all MACAW evaluations were "no", so that the actual P-score (an exponential number) is reported. Prendergast and Ziff reported that all negative controls gave P = 1. For all loop sizes from 4 to 12, a score of P = 1 was obtained.

Playing With Blocks: tRNA Synthases

A   B

		Motif 1					Scrambled set	
ECOLYS	194	IRQFMVNRGFMEVET	P	MM	283	L	RNMFIFTSTLDRV	K
YSTLYS	257	IRRFLDQRKFIEVET	P	MM	42	L	DTIDEAALVYPHV	K
ECOASP	148	VRRFMDDHGFLDIET	P	ML	137	L	AETLRTEFCQNGI	K
RATASP	206	FRETLINKGFVEIQT	P	KI	210	L	RTNLQAKFLAQFG	K
YSTASP	258	FREYLATKKFTEVHT	P	KL	205	L	TFRPTDLNKKESY	K
ECOHIS	29	LKNVLGSYGYSEIRL	P	IV	56	L	ARVHEAWLLQLRL	K
HAMHIS	80	IICCFKRHGAEVIDT	P	VF	276	L	LEKRYAFPGKAKR	K
YSTHIS	79	LSGLFKKHGGVTIDT	P	VF	163	L	KALELGKLLKLSR	K
ECOPHE	132	EI.EDDYHNFDALNI	P	GH	278	L	TRVWAKHIGNDAI	K
MITPHE	91	EPVVTTMENFDSLGF	P	KD	346	L	TKFNQAARDDEEG	K
YSTPHE	253	QYVETGFWNFDALYV	P	QQ	370	L	LAARVASEKTEHL	K
ECOASN	148	LHRFFNEQGFFWVST	P	LI	268	L	LEGAVARKLVVGF	K
YSTTHR	367	LRTEYRKRGYEEVIT	P	NM	479	L	TAISDAEREEMST	K
ECOTHR	**281**	**VRSKLKEYQYQEVKG**	**P**	**FM**	624	L	LLKCKAWTLTQNN	K
ECOPRO	**57**	**VREEMNNAGAIEVSM**	**P**	**VV**	89	L	DNLRGQAAGMAQR	K
ECOSER	**181**	**LDLHTEQHGYSENYV**	**P**	**YL**	306	L	RAVQVAYFPKSVL	K
YSTSER	196	GLQFLAAKGYIPLQA	P	VM	336	L	VHQGKAMCYQLNN	K

Figure 1. Comparison of a block derived from aminoacyl tRNA synthetase sequences to a block constructed from scrambled sequences.

(A) Motif 1 from Figure 3 of Eriani *et al.* (1990). using their criteria for strictly and strongly conserved regions. The sequences in bold type show the initial alignment from their Fig. 2. (B) A block from scrambled sequences with average searching strength found by the MOTIF program (Smith et al.. 1990) in the 17 randomized synthetase sequences. The position of the first residue is indicated for each segment. The single-letter code for the amino acid residues is as follows: A. Ala: C. Cys: D. Asp: E. Glu: F. Phe: G. Gly: H. His: I. Ile: K. Lys: L. Leu: M. Met: N. Asn: P. Pro: Q. Gln: R. Arg: S. Ser: T. Thr: V. Val: W. Trp: and Y. Tyr.

Playing With Blocks:RTase

```
EST1   VFFLDLS..FDFI   57   DLINSPLNCSGNIYSHRPKRSYLFRED  11   LTDFNDDYVY  45   GVIW.NASK
        *  **   *           *         *                         *   * ***      * *    *
    ---Motif A---  17-87  -----------Motif B----------  5-66  --Motif C-  5-62  -Motif D-
        *   *   *            *          *  * **                 *  * ** **     * *    *
CBPB   ASVEDVGLSYEGR   22   DKPII.FIDGGIHAREWIAPSTVTYIV  35   AYTFTDDRLW  13   GSVL..GCK
```

Figure 3. Alignment of EST1 and Carboxypeptidase B (CBPB) with four blocks derived from selected RNA-dependent RNA polymerases using the procedure of Lunblad and Blackburn (1990).

The numbers indicate distance in amino acid residues between the blocks for the known polymerases at the top, and for EST1 and CBPB beneath. The EST1 alignment is taken from their figure. Asterisks indicate conserved residues for both the reverse transcriptases and the RNA polymerases. To align CBPB using the same rules. Motif C was anchored at the [F Y]xDD and the other 3 invariant residues (in bold) were found by scanning the flanking regions while maintaining interblock distances consistent with those seen in known polymerases. A single gap extension was allowed, consistent with their arbitrary extension of the 1-amino acid gap to 2 amino acids in Motif A. Hydrophobic residues are MILVFWAP.

Conclusion

The Best Alignment Method:
- Your Brain
- The Right Data

The Best Evaluation:
- Your Eyes
- Experimental Information (SwissProt)

What Can I Conclude:
- Homology=> Information Extrapolation

How Can I go Further?:
- PrositePatterns.
- PrositeProfiles.