

>ref|NP\_115976.2| caspase recruitment domain-containing protein 6 [Homo sapiens]

MATESTPSEIIERERKKLLEILQHDPDSILDTLTSRRLISEEEYETLENTDLLKKSRLK  
LILVQKKGEATCQHFLKCLFSTFPQSAACGLRHEVLKHENTVPPQSMGASSNSEDASFSP  
GIKQPEAPEITVFFSEKEHLDLETSEFFRDKKTSYRETALSARKNEKEYDTPVTLTSSV  
EKVGCEVPATITTYIKDGQRYEELDDSLYLKGEEYLGSDVTPEDAEATVEEEVYDDPEHVG  
YDGEEDFENSETTEFSGEEPSYEGSETSLSLSEEEQEKSIERKKVFKDVLLCLNMDRSRK  
VLPDFVKQFSLDRGCKWTPESPGLAWNFLMKVQARDVTARDSILSHKVLDEDSKEDLLA  
GVENLEIRDIQTINPLDVLCAATMLCSDSSLQQRQVMSNMYQCQFALPLLLPDAENNKSI  
LGAMKDIVKKQSTQFSGGPTEDTEKFLTLMKMPVISFVRLGYCSFSKSRILNTLLSPAQL  
KLHKIFLHQDLPLLVLPRIQISDGLVEITWCFPDSDDRKEN-P-FFQKPVALANLRGNLES  
FWTQFGFLMEVSSAVFFFTDCLGEKEWDLMLFLGEAAIERCYFVLSSQARESEEAQIFQR  
ILNLKPAQLLFWERGDAGDRRNMEGLQAALQEVMFSSCLRCVSVEDMAALARELGIQVD  
EDFENTQRIQVSSGENMAGTAEGEGQQRHSQKSSSKSQALMPIQEPGTQCELSQNLQNL  
YGTVPVFRPVLNSWLPFTRIGGNFNHVSLSKASWVMGRPFGEQRPKWFHPLPFQNAQAQG  
RGKSFGIQSFHPQIFYSGERFMKFSRVARGCHSNGTFGRLPICQHVQACPERPQMMGT  
LERSRAVASKIGHSYSLDSQPARAVGKPPWQQACTRVTELTATGKLIR-TSHIGKPHPQ  
SFQPAATQK-LRPASQQGVQMKQTGGASNPALQIGSHPMCKSSQFKSD---QSNPSTVK  
HSQPKPFHSPVPSQPKSSQTKSCSQPSQTKPSPCKSTQPKPSQWPPQSKPSQPRPPQPK  
SSSTNPSQAKAHHSKAGQKRGGKH

>ref|XP\_008990374.2| caspase recruitment domain-containing protein 6 isoform X1 [Callithrix jacchus]

MATESTPSEIIERERKKLLEILQHDLDLSILDTLTSRRLISEEEYEALENVTDLLKKSRLK  
LILVQKKGEVTCHEFLKCLFSTFPQSAAVCGLRH-----GKEEYLG  
SVDSPKDAEATV-----EEDYDDPE-----  
-----HAGYDGEEDFKYS-----ETTE  
FSGEE-----PSYEDSQISLSLEEEQEKSMKERKKVFKDVLLCLNMDRSRK  
VLPDFVKQFSLDRGCKWTPESPGLAWNFLMKIQALDVTARDSILRHKVLDEGSKEDLLA  
GVENLEIQDIQTINPLDVICATMLCSDSSLQQRQVMSNMYQCQFALPLLLPDAENNKSI  
LGAMKDIVKKQSTQFSGGPTGDTEKFLTLMKMPVISFVRLGYCSFSKSRILNTLLSPAQV  
KSHRIFLHQDLPLLVLPRIQISDGLIEITWCFPDGDDLKENVS-VFQKPVALANLRGNLES  
FWTQFGFLMEVSSAVFFFTDYLGEKEWDLMLFLGEAAIERCYFVLSPQARESEEAQIFQR  
VLNLKPAQLLFWEGEDAGDRRNIEGLQAALREVMFSSSLRCVSVEDMASLARELGIQVD  
EDFANAQRIQISSGENMAGTAEGESQQRHSQKSSSKSQALMPIQEPGTQCEFSHNLQNL  
YGTVPVFRPLPENSWPLPTRVGGNFNRVSLKAPWVLGCHFGSEQRSKWFCPLPFQNAQAQG  
RGKSFGFQSFHPQRFYSGERFMKFSRSARGCYSNGTFVRPPRPISQRVQACPERPQIMGT  
PERSWAVVFHVGHSCSLGSLQPARAVGKPPWQQACPWGTQLTEATGKPMG-TSHIEKPHPQ  
SFQPAGATQK-LRPASQQGAQKKTQGRPSNPALQIGSHPMCKSPQFKSD---QSNPSQVK  
HSQPKPFHSPVPSQPKSSQTKSSQSPSQTKCSPCKSIQPKPSQPRPPQSKPPQPRPTQPK  
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>ref|XP\_024434398.1| caspase recruitment domain-containing protein 6 [Desmodus rotundus]

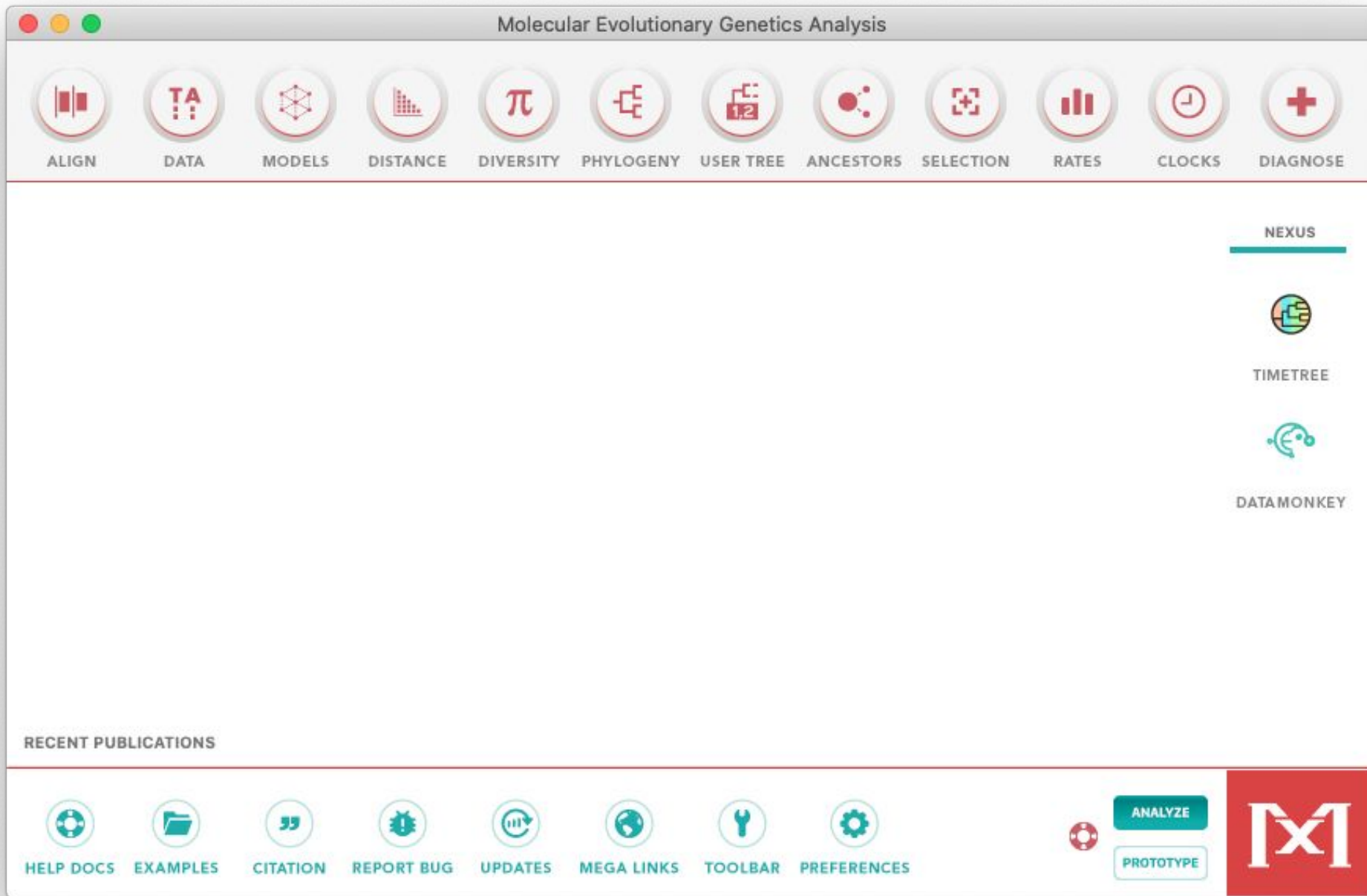
MATGSAPSEIIERERKKLLEVLQDDLDSVLDLSLTSRRLISEDEYETLEDMDPLKKSRLK  
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GEKDPENSELTVPFTEKEHLDLETSETFVDKKTSGEMAWPSRARAKDCHTPAVPLPHSV  
EDVEYEVSKAVHYLQDGQRYDEIDDSLYLGKEDCLESVMYSEDAESAEEEDPSDPEHIV  
YDSEEEPAYSETTEVSQSHGDLTGSLSEEEER-MEEKKKVFKNVLSCLNMDRSRK  
LWPDVAVKQFSLDRGCKWTPETPGDLAWSFLMKVQALDLTAREPTLRPEALRQSGGESPA

## Exemplo de arquivo FASTA

## Proteínas CARD6 de alguns mamíferos

## Adicionar os fragmentos de proteínas

## De *T. cruzi* similares a CARD6



**Abrir o programa MEGA**

**Nesse tutorial usaremos a versão MEGAX**

**Se você estiver usando uma versão diferente pode tentar seguir o tutorial.**

**Normalmente as opções não mudam muito entre versões próximas.**



Edit/Build Alignment

Edit/View Sequencer Files (Trace)...

Open Saved Alignment Session...

Show Web Browser

Query Databanks

Do BLAST Search

Clicar no opção “ALIGN” indicado pela seta azul

Escolher a opção “Edit/Build Alignment” indicada pela seta vermelha

NEXUS



TIMETREE



DATAMONKEY

RECENT PUBLICATIONS



HELP DOCS



EXAMPLES



CITATION



REPORT BUG



UPDATES



MEGA LINKS



TOOLBAR



PREFERENCES



ANALYZE

PROTOTYPE

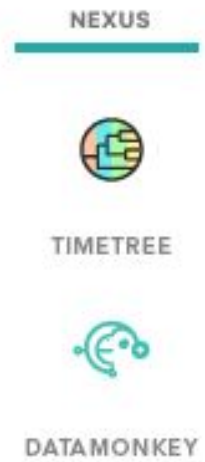
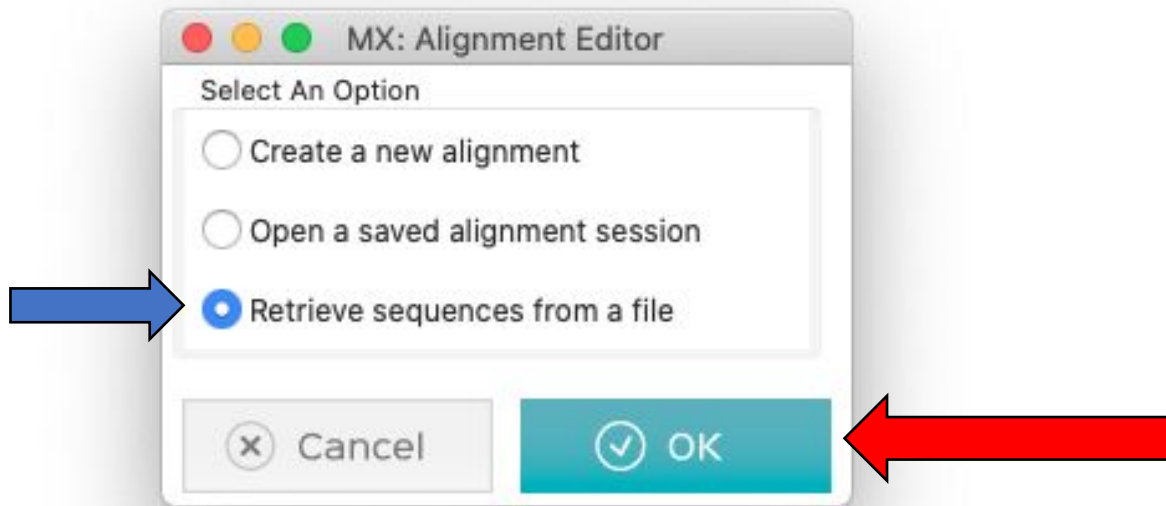




A janela MX: Alignment Editor  
irá aparecer

Escolher “Retrieve sequence  
from a file” indicado pela seta  
azul

Clicar em “OK” indicado pela  
seta vermelha



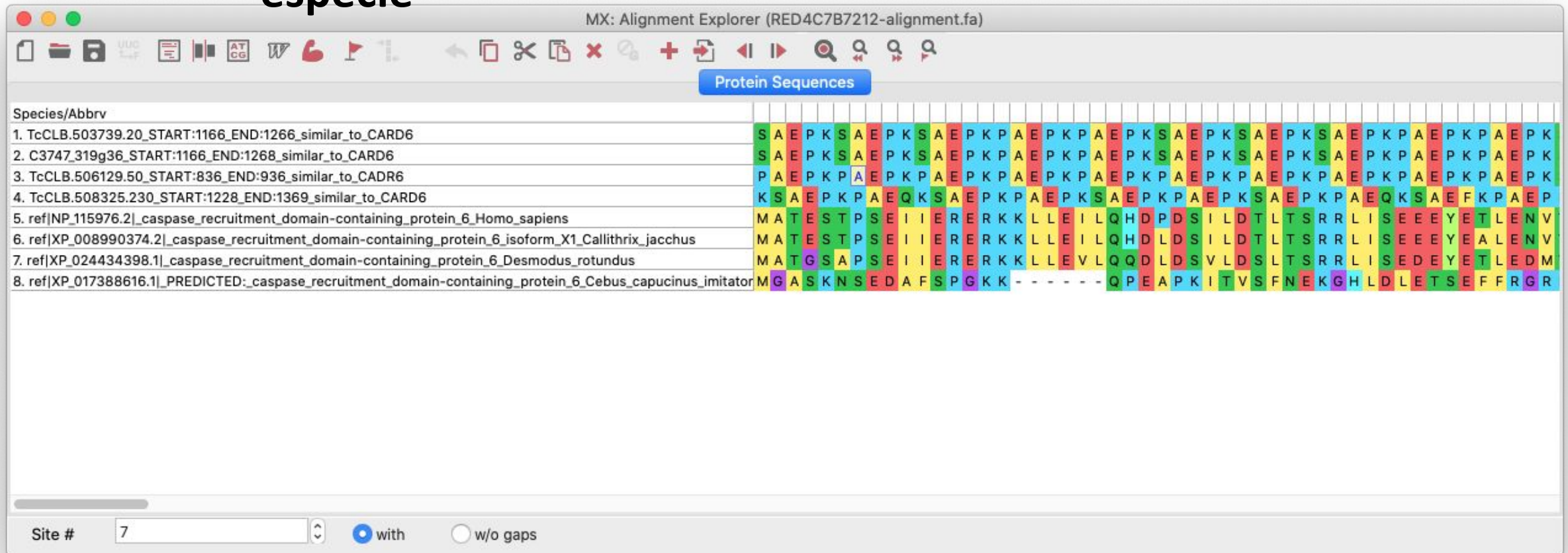
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## A janela MX: Alignment Explorer irá aparecer

## Mostrando em cada linha a sequência de uma espécie



The screenshot shows the MEGAX software interface. The 'Edit' menu is open, displaying various editing options. A blue arrow points to the 'Edit' menu item in the top bar. A red arrow points to the 'Delete Gaps' option in the 'Edit' menu. A yellow arrow points to the 'Select All' option in the 'Edit' menu. The background shows a sequence alignment view with a 'Protein Sequences' button and a list of sequences. The bottom of the interface shows a 'Site #' field with the value '7' and radio buttons for 'with' and 'w/o gaps'.

**MEGAX** Data **Edit** Search Alignment Web Sequencer Display Help

MX: Alignment Explorer (RED4C7B7212-alignment.fa)

Protein Sequences

Species/Abbrv

1. TcCLB.503739.20\_S

2. C3747 319a36 STA

3. [REDACTED]

4. TcCLB.508325.230

5. ref|NP\_115976.2|\_ca

6. ref|XP\_008990374.

7. ref|XP\_024434398.

8. ref|XP\_017388616.1

Undo ⌘Z

Copy ⌘C

Cut ⌘X

Paste ⌘V

Delete

Delete Gaps ⌘⌘

Insert Blank Sequence ⌘N

Insert Sequence From File ⌘I

Select Site(s)

Select Sequence(s)

Select All ⌘A

✓ Allow Base Editing

Modify All Bases To Upper Case

Site # 7 with w/o gaps

**Antes de fazer o alinhamento vamos remover os gaps que podem estar presentes em sequencias previamente alinhadas**

**Clicar em “Edit” indicado pela seta azul**

**Depois escolher a opção “Select All”**

**Novamente clicar em “Edit” indicado pela seta azul**

**Depois escolher a opção “Delete Gaps” indicado pela seta vermelha**

MEGAX Data Edit Search Alignment Web Sequencer Display Help

MX: Alignment Explorer (RED4C7B7212-alignment.fa)

Align Protein Protein Sequences

Species/Abbrv

1. TcCLB.503739.20_START:1166_END:1266_similar_to_CARD6	S	A	E	P	K	S	A	E	P	K	S	A	E	P	K	P	A	E	P	K	P	A	E	P	K	S
2. C3747_319g36_START:1166_END:1268_similar_to_CARD6	S	A	E	P	K	S	A	E	P	K	S	A	E	P	K	P	A	E	P	K	P	A	E	P	K	S
3. TcCLB.506129.50_START:836_END:936_similar_to_CADR6	P	A	E	P	K	P	A	E	P	K	P	A	E	P	K	P	A	E	P	K	P	A	E	P	K	P
4. TcCLB.508325.230_START:1228_END:1369_similar_to_CARD6	K	S	A	E	P	K	P	A	E	Q	K	S	A	E	P	K	P	A	E	P	K	S	A	E	P	K
5. ref NP_115976.2 _caspase_recruitment_domain-containing_protein_6_Homo_sapiens	M	A	T	E	S	T	P	S	E	I	I	E	R	E	R	K	K	L	L	E	I	L	Q	H	D	P
6. ref XP_008990374.2 _caspase_recruitment_domain-containing_protein_6_isoform_X1_Callithrix_jacchus	M	A	T	E	S	T	P	S	E	I	I	E	R	E	R	K	K	L	L	E	I	L	Q	H	D	L
7. ref XP_024434398.1 _caspase_recruitment_domain-containing_protein_6_Desmodus_rotundus	M	A	T	G	S	A	P	S	E	I	I	E	R	E	R	K	K	L	L	E	V	L	Q	Q	D	L
8. ref XP_017388616.1 _PREDICTED:_caspase_recruitment_domain-containing_protein_6_Cebus_capucinus_imitator	M	G	A	S	K	N	S	E	D	A	F	S	P	G	K	K	Q	P	E	A	P	K	I	T	V	S

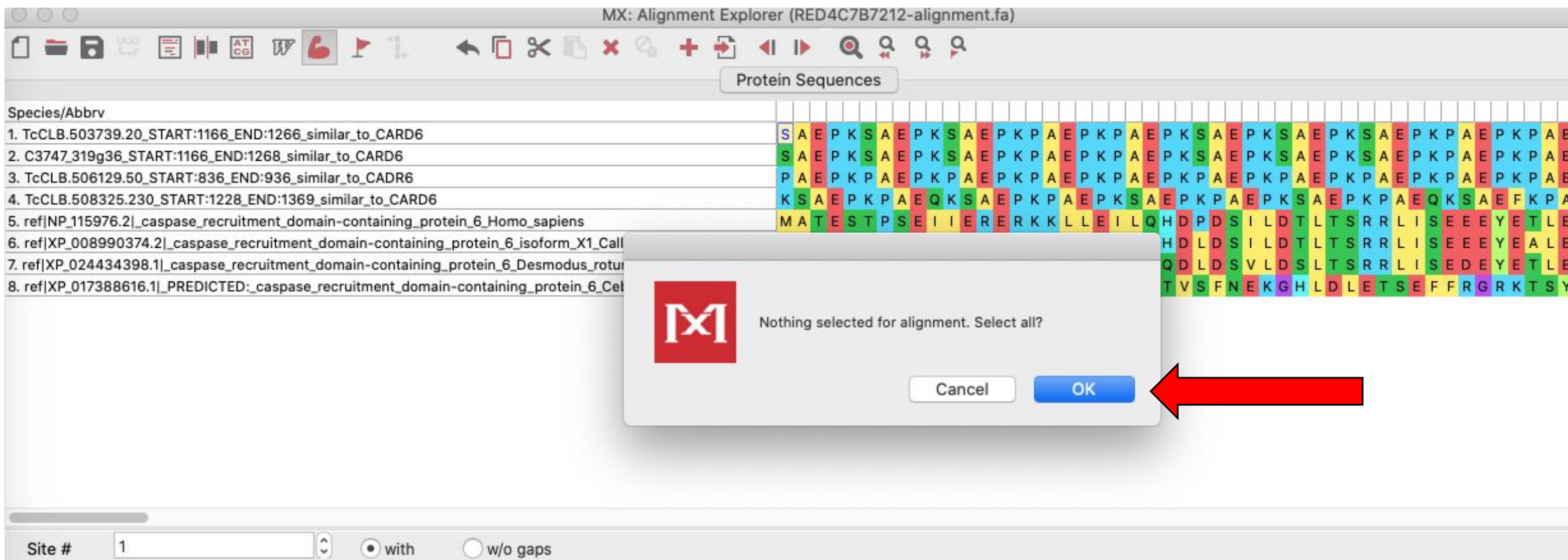
Site # 1 ☐ with ☐ w/o gaps

Para fazer o alinhamento

Clique no ícone do algoritmo MUSCLE (braço) indicado pela seta azul

Depois em “Align Protein” indicado pela seta vermelha

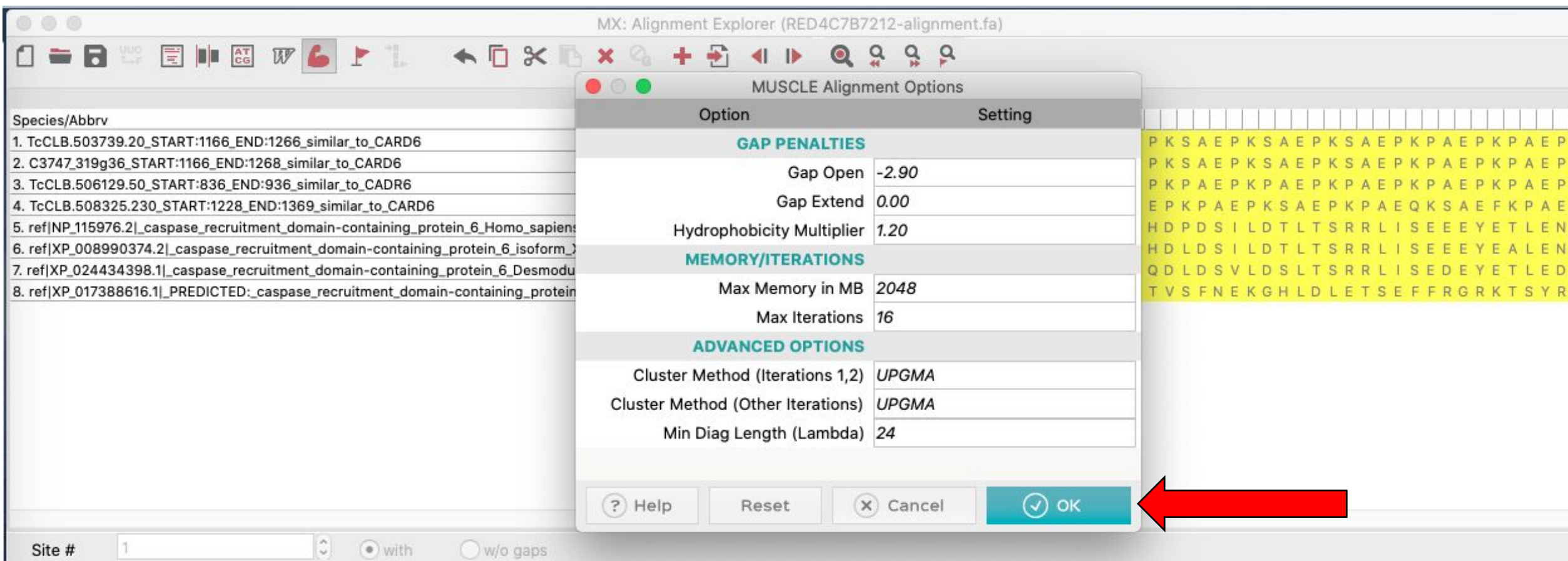




**O programa MEGA irá informar que nenhuma sequência está selecionada e perguntar se você quer selecionar todas.**

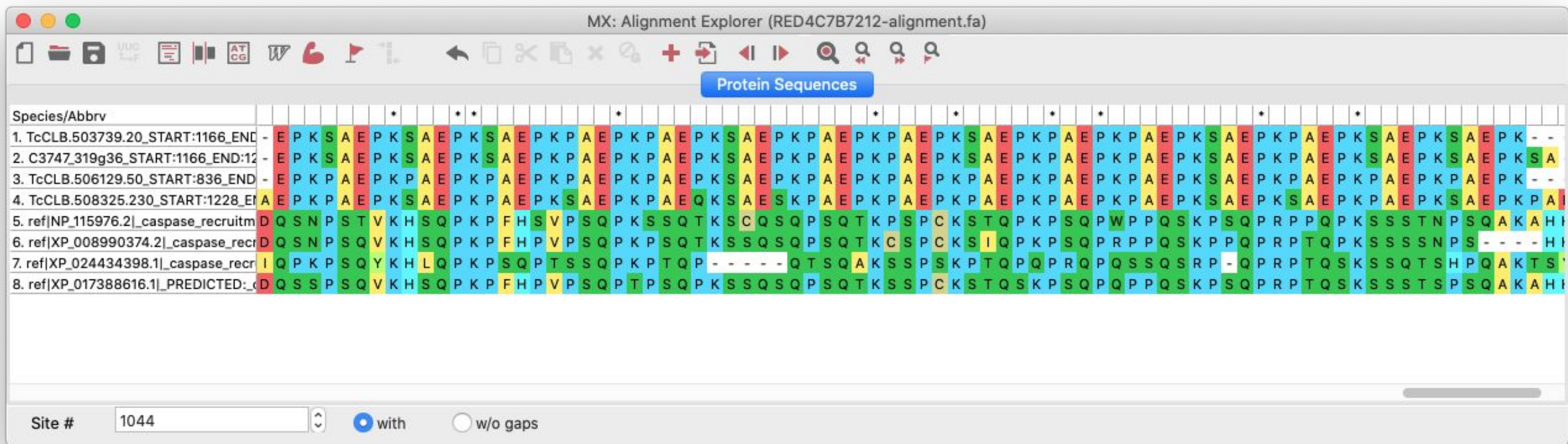
**Queremos alinhar todas, então clique em “OK” indicado pela seta em vermelho**





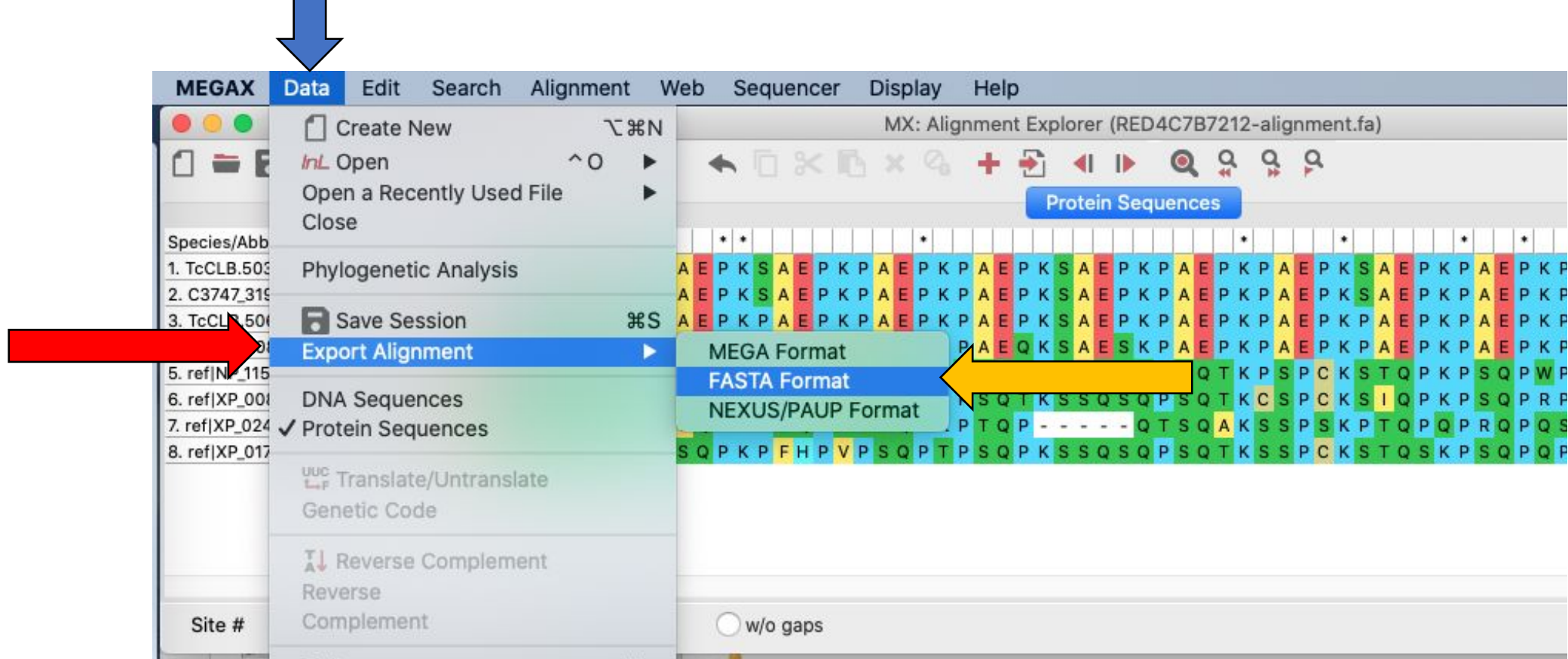
A janela “MUSCLE Alignment Options” irá abrir.

Não vamos alterar nenhum dos parâmetros de alinhamento do programa. Clique em “OK” indicado pela seta em vermelho. Em seguida o MUSCLE irá alinhar suas sequências.



**O resultado do alinhamento aparece na janela “MX: Alignment Explorer” irá abrir.**

**Vamos procurar por trechos de similaridade ao longo do alinhamento.**



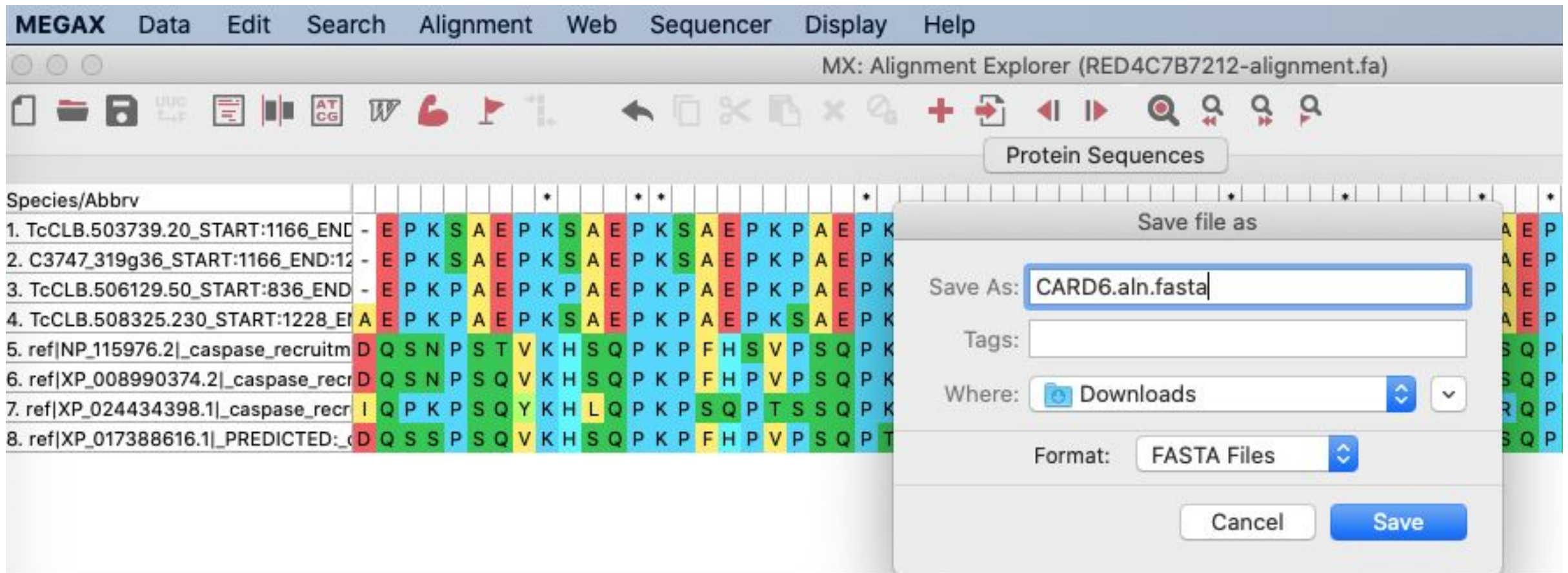
Vamos exportar esse alinhamento no formato FASTA.

Clicar em “Data” indicado pela seta azul

Escolher “Export Alignment” indicado pela seta vermelha

Clicar em “FASTA format” indicado pela seta laranja





Sugiro colocar o nome do gene (CARD6) seguido da extensão “aln.fasta” para indicar que são sequencia já alinhadas e no formato fasta

CARD6.aln.fasta