

Jimmi Nguyen A15976796

BIMM 143

Professor Grant

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Find a Gene Assignment Part 1

[Q1] Tell me the name of a protein you are interested in. Include the species and the accession number. This can be a human protein or a protein from any other species as long as it's function is known. If you do not have a favorite protein, select human RBP4 or KIF11. Do not use beta globin as this is in the worked example report that I provide you with online.

Name: Tumor protein p53

Accession: NP_000537

Species: Homo Sapiens

[Q2] Perform a BLAST search against a DNA database, such as a database consisting of genomic DNA or ESTs. The BLAST server can be at NCBI or elsewhere. Include details of the BLAST method used, database searched and any limits applied (e.g. Organism).

Method: TBLASTN search against chicken ESTs

Database: Expressed Sequence Tags (est)

Organism: Chicken (Taxid:9031)

Translated BLAST: tblastn

blastn blastp blastx **tblastn** tblastx

TBLASTN search translated nucleotide databases using a protein query

Enter Query Sequence

Enter accession number(s), gi(s), or FASTA sequence(s) [?](#) [Clear](#)

>NP_000537.3 cellular tumor antigen p53 isoform a [Homo sapiens]
MEEPQSDPSVEPPLSQETFSDLWKLLPENNVLSPLPSQAMDDLMLSPDDI
EQWFTEDPGPDEAPRMPEAA
PPVAPAPAAPTPAAPAPAPSWPLSSSVPSQKTYQGSYGFRLLGFLHSGTARS

Query subrange [?](#)

From

To

Or, upload file No file chosen [?](#)

Job Title
Enter a descriptive title for your BLAST search [?](#)

☐ Align two or more sequences [?](#)

Choose Search Set

Database [?](#)

Organism Optional ☐ exclude
Enter organism common name, binomial, or tax id. Only 20 top taxa will be shown [?](#)

Exclude Optional ☐ Models (XM/XP) ☐ Uncultured/environmental sample sequences

Limit to Optional ☐ Sequences from type material

Entrez Query Optional
Enter an Entrez query to limit search [?](#) [YouTube](#) [Create custom database](#)

BLAST Search database est using Tblastn (search translated nucleotide databases using a protein query)
☒ Show results in a new window

Your search is limited to records that include: chicken (taxid:9031)

Job Title

NP_000537.3 cellular tumor antigen p53 isoform...

RID

Y2HV/KW3K016 Search expires on 02-08 01:31 am [Download All](#)

Program

TBLASTN [Citation](#)

Database

est [See details](#)

Query ID

lcl|Query_20167

Description

NP_000537.3 cellular tumor antigen p53 isoform a [Homo ...

Molecule type

amino acid

Query Length

393

Other reports

[?](#)

Filter Results

Organism only top 20 will appear ☐ exclude

Type common name, binomial, taxid or group name

[+ Add organism](#)

Percent Identity

to

E value

to

Query Coverage

to

Filter

Reset

Descriptions

Graphic Summary

Alignments

Taxonomy

Sequences producing significant alignments

Download

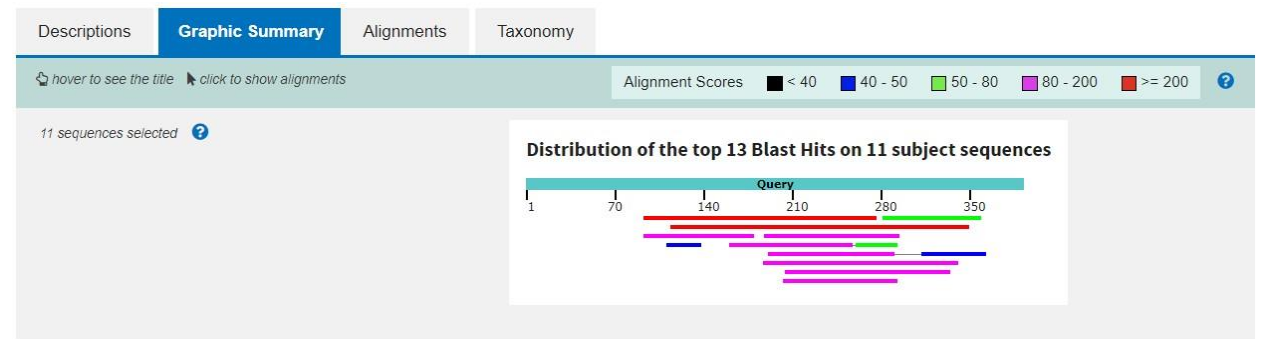
Select columns

Show 100

☒ select all 11 sequences selected

[GenBank](#)
[Graphics](#)

	Description	Scientific Name	Max Score	Total Score	Query Cover	E value	Per. Ident	Acc. Len	Accession
<input checked="" type="checkbox"/>	pgr1n.pk002.117 Normalized Chicken Reproductive Tract cDNA Library (pgr1n) Gallus gallus cDNA clone pgr1n.pk002.117	Gallus gallus	268	268	46%	1e-88	68.11%	568	BM440292.1
<input checked="" type="checkbox"/>	603101519F1.CSEQCHN03 Gallus gallus cDNA clone ChEST34e5'5'' mRNA sequence	Gallus gallus	246	246	60%	9e-79	50.21%	854	BU203323.1
<input checked="" type="checkbox"/>	pat.pk0040.d2.f1 chicken activated T cell cDNA Gallus gallus cDNA clone pat.pk0040.d2.f5' similar to cellular tumor anti	Gallus gallus	174	174	27%	6e-52	76.64%	547	AI981042.1
<input checked="" type="checkbox"/>	pgr1n.UA001.559 Normalized chicken reproductive tract (pgr1n) Gallus gallus cDNA clone UAGgRT21_D09_03061705...	Gallus gallus	166	217	33%	1e-49	77.78%	408	DT659470.1
<input checked="" type="checkbox"/>	pqp2n.pk008.m15 Normalized chicken pituitary/hypothalamus/pineal cDNA library (pqp2n) Gallus gallus cDNA clone pqp2n.pk008.m15	Gallus gallus	169	216	38%	1e-49	79.00%	639	CD215735.1



[Download](#) ▾ [GenBank](#) [Graphics](#) [▼ Next](#) [▲ Previous](#) [◀ Descriptions](#)

pgr1n.pk002.i17 Normalized Chicken Reproductive Tract cDNA Library (pgr1n) Gallus gallus cDNA clone pgr1n.pk002.i17 5' similar to sp|P10360|P53_CHICK CELLULAR TUMOR ANTIGEN P53 (TUMOR SUPPRESSOR P53) pir|S02193 cellular tumor antigen p53 - chicken emb|CAA31456.1| (X13057) nuclear protein p53 (AA 1 - 367) [Gallus gallus], mRNA sequence

Sequence ID: [BM440292.1](#) Length: **568** Number of Matches: **1**

Range 1: 13 to 567 [GenBank](#) [Graphics](#) [▼ Next Match](#) [▲ Previous Match](#)

Score	Expect	Method	Identities	Positives	Gaps	Frame
268 bits(685)	1e-88	Compositional matrix adjust.	126/185(68%)	147/185(79%)	1/185(0%)	+1
Query 94	SSSVPSQTKYQSGYGRFLGFLHSGTAKSVTCTYSPALNKMFQQLAKTCPVQLWVDSTPPP	153				
Sbjct 13	S VPS + Y G + FR+GF+ +GTAKSVTCTYSP LNK++C+LAK CPVQ+ V PPP	192				
Query 154	GTRVRAMAIYKQSQHMTVEVRRCPHHERCSD-SDGLAPQHLIRVEGNLRVEYLDNRNTF	212				
Sbjct 193	G+ +RA+A+YK+S+H+ EVVRRCPHHERC +DGLAP QHLIRVEGN + Y DD T	372				
Query 213	RHSVVVPYEPPEVSGDCTTIHNYMNCNSSCMGMMNRRPILTIITLEDSSGNLGRNSFEV	272				
Sbjct 373	RHSVVVPYEPPEVSGDCTT+ YN+MNCNSSCMGMMNRRPILTI+TL G LLGR	552				
Query 273	RVCAC 277					
Sbjct 553	RVCAC 567					

Alignment Details:

pgr1n.pk002.117 Normalized Chicken Reproductive Tract cDNA Library (pgr1n) Gallus gallus cDNA clone pgr1n.pk002.117 5' similar to sp|P10360|P53_CHICK CELLULAR TUMOR ANTIGEN P53 (TUMOR SUPPRESSOR P53) pir|S02193 cellular tumor antigen p53 - chicken emb|CAA31456.1| (X13057) nuclear protein p53 (AA 1 - 367) [Gallus gallus], mRNA sequence

Sequence ID: [BM440292.1](#) Length: 568 Number of Matches: 1

Range 1: 13 to 567 [GenBankGraphics](#) [Next Match](#) [Previous Match](#)

Alignment statistics for match #1

Score	Expect	Method	Identities	Positives	Gaps
268 bits(685)	1e-88	Compositional matrix adjust.	126/185(68%)	147/185(79%)	1/185(0%)

Query 94 SSSVPSQKTYQGSYGFRGLHSGTAKSVTCTYSPALNKMFCQLAKTCPVQLWVDSTPPP 153

S VPS + Y G + FR+GF+ +GTAKSVTCTYSP LNK++C+LAK CPVQ+ V PPP

Sbjct 13 SPVVPSTEDYGGDFDFRVGFVEAGTAKSVTCTYSPVLNKVYCR LAKPCPVQVRVGVAPPP 192

Query 154 GTRVRAMAIYKQSQHMTEVVRRCPPHHERCSD-SDGLAPPQH LIRVEGNLRVEYLDDRNTF 212

G+ +RA+A+YK+S+H+ EVVRRCPHHERC +DGLAP QHLIRVEGN + Y DD T

Sbjct 193 GSSLRAVAVYKKSEHVAE VVRRCPHHERCGGGTDGLAPA QHLIRVEGNPQARYHDDETTK 372

Query 213 RHSVVPYPPEVGS DCTTIHYNM CNSSCMGGMNRRPILTIITLEDSSGNLLGRNSFEV 272

RHSVVPYPPEVGS DCTT+ YN+MCNSSCMGGMNRRPILTI+TL G LLGR

Sbjct 373 RHSVVPYPPEVGS DCTTVLYNFM CNSSCMGGMNRRPILTILTLXXPGGQLLGRR CXXX 552

Query 273 RVCAC 277

RVCAC

Sbjct 553 RVCAC 567

[Q3] Gather information about this “novel” protein. At a minimum, show me the protein sequence of the “novel” protein as displayed in your BLAST results from [Q2] as FASTA format (you can copy and paste the aligned sequence subject lines from your BLAST result page if necessary) or translate your novel DNA sequence using a tool called EMBOSS Transeq at the EBI. Don’t forget to translate all six reading frames; the ORF (open reading frame) is likely to be the longest sequence without a stop codon. It may not start with a methionine if you don’t have the complete coding region. Make sure the sequence you provide includes a header/subject line and is in traditional FASTA format.

Chosen Sequence:

```
>BM440292.1_1 pgr1n.pk002.117 Normalized Chicken Reproductive Tract cDNA Library
(pgr1n) Gallus gallus cDNA clone pgr1n.pk002.117 5' similar to sp|P10360|P53_CHICK
CELLULAR TUMOR ANTIGEN P53 (TUMOR SUPPRESSOR P53) pir|S02193 cellular tumor
antigen p53 - chicken emb|CAA31456.1| (X13057) nuclear protein p53 (AA 1 - 367) [Gallus
gallus], mRNA sequence
RAAPSPVVPSTEDYGGDFDFRVGFVEAGTAKSVTCTYSPVLNKKVYCRLAKPCPVQVRV
GV
APPPGSSLRAVAVYKKSEHVAEVVRRCPHHERCGGGTDGLAPAQHLIRVEGNPQARYH
DD
ETTKRHSVVVPYEPPEVGSDCTTVLYNFMCNSSCMGGMNRRPILTILTLXXPGGQLLGR
R
CXXXRVCACX
```

Name: Cellular Tumor Antigen P53

Species: Gallus gallus

Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi;
Archelosauria; Archosauria; Dinosauria; Saurischia; Theropoda;
Coelurosauria; Aves; Neognathae; Galloanserae; Galliformes;
Phasianidae; Phasianinae; Gallus.

[Q4] Prove that this gene, and its corresponding protein, are novel. For the purposes of this project, “novel” is defined as follows. Take the protein sequence (your answer to [Q3]), and use it as a query in a blastp search of the nr database at NCBI.

- If there is a match with 100% amino acid identity to a protein in the database, from the same species, then your protein is NOT novel (even if the match is to a protein with a name such as “unknown”). Someone has already found and annotated this sequence, and assigned it an accession number.
- If the top match reported has less than 100% identity, then it is likely that your protein is novel, and you have succeeded.
- If there is a match with 100% identity, but to a different species than the one you started with, then you have likely succeeded in finding a novel gene.
- If there are no database matches to the original query from [Q1], this indicates that you have partially succeeded: yes, you may have found a new gene, but no, it is not actually homologous to the original query. You should probably start over.

Details:

A BLASTP search against NR database yielded a top hit result to a protein from Gallus gallus. See additional screen shots below for top hits and selected alignment details:

Enter Query Sequence

Enter accession number(s), gi(s), or FASTA sequence(s) [?](#) [Clear](#)

Query subrange [?](#)

From
To

GNPQARYHDD

ETTKRHSVVVPYEPPEVGSDDCTTVLYNFMCSNCGMNRRLTILTLXX

PGGQLLR

CXXXRVACAX

Or, upload file

No file chosen [?](#)

Job Title

BM440292.1_1_pgr1n.pk002.117 Normalized Chicken...

Enter a descriptive title for your BLAST search [?](#)

☐ Align two or more sequences [?](#)

Choose Search Set

Databases

☒ Standard databases (nr etc.): New
☐ Experimental databases

Compare

☐ Select to compare standard and experimental database [?](#)

Standard

Database

Non-redundant protein sequences (nr) [?](#)

Organism
Optional

☐ exclude

Enter organism common name, binomial, or tax id. Only 20 top taxa will be shown [?](#)

Exclude
Optional

☐ Models (XM/XP)
☐ Non-redundant RefSeq proteins (WP)
☐ Uncultured/environmental sample sequences

Program Selection

Algorithm

☐ Quick BLASTP (Accelerated protein-protein BLAST)
☒ blastp (protein-protein BLAST)
☐ PSI-BLAST (Position-Specific Iterated BLAST)
☐ PHI-BLAST (Pattern Hit Initiated BLAST)
☐ DELTA-BLAST (Domain Enhanced Lookup Time Accelerated BLAST)

Choose a BLAST algorithm [?](#)

Search database nr using Blastp (protein-protein BLAST)
☒ Show results in a new window

The top result is to a protein from Gallus gallus (chicken), see second screen shot below for alignment details:

Sequences producing significant alignments									
<input checked="" type="checkbox"/> select all 100 sequences selected									
GenPept Graphics Distance tree of results Multiple alignment MSA Viewer									
	Description	Scientific Name	Max Score	Total Score	Query Cover	E value	Per. Ident	Acc. Len	Accession
<input checked="" type="checkbox"/>	cellular tumor antigen p53 [Gallus gallus]	Gallus gallus	378	378	99%	2e-129	97.35%	367	NP_990595.1
<input checked="" type="checkbox"/>	cellular tumor antigen p53 isoform X3 [Anas platyrhynchos]	Anas platyrhyn...	335	335	96%	1e-112	87.98%	352	XP_038027747.1
<input checked="" type="checkbox"/>	cellular tumor antigen p53 isoform X1 [Anas platyrhynchos]	Anas platyrhyn...	335	335	96%	1e-112	87.98%	365	XP_038027745.1
<input checked="" type="checkbox"/>	cellular tumor antigen p53 isoform X2 [Anas platyrhynchos]	Anas platyrhyn...	335	335	96%	1e-112	87.98%	364	XP_038027746.1
<input checked="" type="checkbox"/>	cellular tumor antigen p53 isoform X2 [Camarhynchus parvulus]	Camarhynchus ...	331	331	98%	7e-112	84.57%	303	XP_030825917.1
<input checked="" type="checkbox"/>	cellular tumor antigen p53 isoform X1 [Accipiter gentilis]	Accipiter gentilis	331	331	94%	2e-111	88.89%	319	XP_049648596.1
<input checked="" type="checkbox"/>	cellular tumor antigen p53 isoform X2 [Accipiter gentilis]	Accipiter gentilis	330	330	94%	3e-111	88.89%	318	XP_049648597.1

cellular tumor antigen p53 [Gallus gallus]

Sequence ID: [NP_990595.1](#) Length: 367 Number of Matches: 1
[See 2 more title\(s\)](#) [See all Identical Proteins\(IPG\)](#)

Range 1: 75 to 263 [GenPept](#) [Graphics](#) [Next Match](#) [Previous Match](#)

Score	Expect	Method	Identities	Positives	Gaps
378 bits(971)	2e-129	Compositional matrix adjust.	184/189(97%)	184/189(97%)	0/189(0%)
Query 1	RAAPSPVVPSTEDYGGDFDFRVGFVEAGTAKSVTCTYSPVLNKVYCRLAKPCPVQVRVGV				60
Sbjct 75	RAAPSPVVPSTEDYGGDFDFRVGFVEAGTAKSVTCTYSPVLNKVYCRLAKPCPVQVRVGV				134
Query 61	APPPGSSSLRAVAVYKKSEHVAEVVRRCPHHERCGGGTDGLAPAQHLIRVEGNPQARYHDD				120
Sbjct 135	APPPGSSSLRAVAVYKKSEHVAEVVRRCPHHERCGGGTDGLAPAQHLIRVEGNPQARYHDD				194
Query 121	ETTKRHSVVVPYEPPEVGSDDCTTVLYNFCNSSCMGGMNRRPILITLTLXXPGGQLLGR				180
Sbjct 195	ETTKRHSVVVPYEPPEVGSDDCTTVLYNFCNSSCMGGMNRRPILITLTLXPGGQLLGR				254
Query 181	CXXXRVAC 189				
	C RVCAC				
Sbjct 255	CFEVRVCAC 263				

Related Information
[Gene](#) - associated gene details
[Identical Proteins](#) - Identical proteins to NP_990595.1

cellular tumor antigen p53 isoform X3 [Anas platyrhynchos]

Sequence ID: [XP_038027747.1](#) Length: 352 Number of Matches: 1

Range 1: 75 to 257 [GenPept](#) [Graphics](#) [Next Match](#) [Previous Match](#)

Score	Expect	Method	Identities	Positives	Gaps
335 bits(859)	1e-112	Compositional matrix adjust.	161/183(88%)	169/183(92%)	0/183(0%)
Query 7	VVPSTEDYGGDFDFRVGFVEAGTAKSVTCTYSPVLNKVYCRLAKPCPVQVRVGVAPPPGS				66
Sbjct 75	MVPSTEDYGGHYDFQLGFQETGTAKSVTCTYSPVLNKLYCRLAKPCPVQVRVGAAPPPGA				134
Query 67	SLRAVAVYKKSEHVAEVVRRCPHHERCGGGTDGLAPAQHLIRVEGNPQARYHDETTKRH				126
Sbjct 135	VLRAVAVYKKSEHVAEVVRRCPHHERNGEGTDGLAPAQHLIRVEGNPQARYHDETTKRH				194
Query 127	SVVVPYEPPEVGSDDCTTVLYNFCNSSCMGGMNRRPILITLTLXXPGGQLLGRRCXXRV				186