

Using BLAST for sequence similarity search

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Topics to be addressed - Survey Result



https://blast.ncbi.nlm.nih.gov/Blast.cgi

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Basic Local Alignment Search Tool

BLAST finds regions of similarity between biological sequences. The program compares nucleotide or protein sequences to sequence databases and calculates the statistical significance. Learn more

Search Betacoronavirus Database

We have created a new BLAST database focused on the SARS-CoV-2 (Severe acute respiratory syndrome coronavirus 2) Sequences. For further detail please visit

NCBI GenBank.

Mon, 03 Feb 2020 10:00:00 EST

More BLAST news...

Web BLAST









BLAST Genomes

Search Enter organism common name, scientific name, or tax id Human Mouse Microbes

Standalone and API BLAST







Downloading the standalone package of executables



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BLAST+ executables

Do you have difficulties running high volume BLAST searches? Do you have proprietary sequence data to search and cannot use the NCBI BLAST web site? Do you have your own research pipeline? Have security or IP concerns about sending searches outside of your organization? If you answered yes to any of these questions, read on!

The NCBI provides a suite of command-line tools to run BLAST called BLAST+. This allows users to perform BLAST searches on their own server without size, volume and database restrictions. BLAST+ can be used with a command line so it can be integrated directly into your workflow.

What are the next steps?



Download and install BLAST+. Installers and source code are available from ftp://ftp.ncbi.nlm.nih.gov/blast/executables/blast+/LATEST/. Download the database you need (see database section below), or create your own. Start searching

For more details, please see the BLAST+ user manual, the BLAST Help manual, the BLAST releases notes, and the article in BMC Bioinformatics (PubMed link). See our versioning policy.

The BLAST+ suite is the currently supported package. The older C toolkit executables are no longer supported. See our versioning policy.

We are always listening and welcome your feedback at BLAST Support Center

Index of /blast/executables/blast+/LATEST/

[parent directory]

Name	Size	Date Modified
ChangeLog	85 B	04/12/2019, 02:52:00
ncbi-blast-2.10.0+-4.src.rpm	19.4 MB	04/12/2019, 02:50:00
ncbi-blast-2.10.0+-4.src.rpm.md5	63 B	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-4.x86_64.rpm	175 MB	04/12/2019, 02:50:00
ncbi-blast-2.10.0+-4.x86_64.rpm.md5	66 B	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-src.tar.gz	24.4 MB	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-src.tar.gz.md5	64 B	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-src.zip	28.5 MB	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-src.zip.md5	61 B	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-win64.exe	86.6 MB	04/12/2019, 02:49:00
ncbi-blast-2.10.0+-win64.exe.md5	63 B	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-x64-linux.tar.gz	222 MB	04/12/2019, 02:52:00
ncbi-blast-2.10.0+-x64-linux.tar.gz.md5	70 B	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-x64-macosx.tar.gz	141 MB	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-x64-macosx.tar.gz.md5	71 B	04/12/2019, 02:53:00
ncbi-blast-2.10.0+-x64-win64.tar.gz	86.3 MB	04/12/2019, 02:50:00
ncbi-blast-2.10.0+-x64-win64.tar.gz.md5	70 B	04/12/2019, 02:53:00
ncbi-blast-2.10.0+.dmg	143 MB	04/12/2019, 02:52:00
ncbi-blast-2.10.0+.dmg.md5	57 B	04/12/2019, 02:53:00

Running BLAST executables

Formatting DBs

\$ makeblastdb -in targetDB.fasta -dbtype [nucl|prot]

Running a regular blastn analysis

\$ blastn -task blastn -query file1.fasta -db targetDB -out file1--vs--targetDB.BlastN.txt

Changing parameters for a more sensitive search and reporting only the top10 hits

\$ blastn -task blastn -query file1.fasta -db targetDB -out file1--vs--targetDB.BlastN-F-W7-1e-3top10.txt -dust no -word_size 7 -evalue 1e-3 -num_descriptions 10 -num_alignments 10 -num_threads 12

Same as above, but generating a tabular output file rather than the long default output format

\$ blastn -task blastn -query file1.fasta -db targetDB -out file1--vs--targetDB.BlastN-F-W7-1e-3top10.tsv -dust no -word_size 7 -evalue 1e-3 -num_descriptions 10 -num_alignments 10 -outfmt [6|7] num_threads 12

Running remotely (using NCBI nr or nt databases), just add the "-remote" option at the end of the cmd line

\$ nohup blastp -query file1.fasta -db nr -out test10-vs-nr.BlastP_remote-top5.txt -num_descriptions 5
-num_alignments 5 -remote &

NOTE: nt.nal or nr.pal file must exist in the current directory, for either non-redundant nucleotide or protein DBs, respectively # They can be obtained by "untaring" one of the n[tr].##.tar.gz files from the ftp://ftp.ncbi.nlm.nih.gov/blast/db/

Tweaking tabular output format options

```
*** Formatting options
-outfmt <String>
 alignment view options:
   0 = Pairwise,
   1 = Query-anchored showing identities,
   2 = Query-anchored no identities,
   3 = Flat query-anchored showing identities,
   4 = Flat query-anchored no identities,
   5 = BLAST XML.
   6 = Tabular,
   7 = Tabular with comment lines,
   8 = Segalign (Text ASN.1),
   9 = Segalign (Binary ASN.1),
  10 = Comma-separated values,
  11 = BLAST archive (ASN.1),
  12 = Segalign (JSON),
  13 = Multiple-file BLAST JSON,
  14 = Multiple-file BLAST XML2,
  15 = Single-file BLAST JSON,
  16 = Single-file BLAST XML2,
  18 = Organism Report
```

<u>NOTE:</u> One may see those instructions by typing any of the blast executable commands (blastn, blastp, blastx, tblastn, or tblastx) followed by "-help"

```
Options 6, 7 and 10 can be additionally configured to produce
a custom format specified by space delimited format specifiers.
The supported format specifiers are:
         qseqid means Query Seq-id
            ggi means Query GI
           qacc means Query accesion
        qaccver means Query accesion.version
           glen means Query sequence length
         ssegid means Subject Seg-id
      sallsegid means All subject Seg-id(s), separated by a ';'
            sgi means Subject GI
         sallgi means All subject GIs
           sacc means Subject accession
        saccver means Subject accession.version
        sallacc means All subject accessions
           slen means Subject sequence length
         gstart means Start of alignment in query
           qend means End of alignment in query
         sstart means Start of alignment in subject
           send means End of alignment in subject
           gseg means Aligned part of query sequence
           sseq means Aligned part of subject sequence
         evalue means Expect value
       bitscore means Bit score
          score means Raw score
         length means Alignment length
         pident means Percentage of identical matches
         nident means Number of identical matches
       mismatch means Number of mismatches
       positive means Number of positive-scoring matches
        gapopen means Number of gap openings
           gaps means Total number of gaps
           ppos means Percentage of positive-scoring matches
         frames means Query and subject frames separated by a '/'
         gframe means Query frame
         sframe means Subject frame
           btop means Blast traceback operations (BTOP)
         staxid means Subject Taxonomy ID
       ssciname means Subject Scientific Name
       scomname means Subject Common Name
      sblastname means Subject Blast Name
      sskingdom means Subject Super Kingdom
        staxids means unique Subject Taxonomy ID(s), separated by a ';'
                       (in numerical order)
      sscinames means unique Subject Scientific Name(s), separated by a ';
      scomnames means unique Subject Common Name(s), separated by a ';'
      sblastnames means unique Subject Blast Name(s), separated by a ';'
                       (in alphabetical order)
     sskingdoms means unique Subject Super Kingdom(s), separated by a ';'
                       (in alphabetical order)
         stitle means Subject Title
     salltitles means All Subject Title(s), separated by a '<>'
        sstrand means Subject Strand
          qcovs means Query Coverage Per Subject
        qcovhsp means Query Coverage Per HSP
        gcovus means Ouery Coverage Per Unique Subject (blastn only)
When not provided, the default value is:
 'qaccver saccver pident length mismatch gapopen qstart qend sstart send
evalue bitscore', which is equivalent to the keyword 'std'
Default = 0'
```

Important filtering options for better results' interpretation

```
#Percent identity
-perc_identity <Real, 0..100>
#Percent query coverage per hsp
-qcov_hsp_perc
<Real, 0..100>
```

#NOTE: Another straightforward alternative is being quite loose (less stringent) on a first BLAST run, and then filtering a large tabular output file with the "awk" bash command, setting desired thresholds for both columns 3 and 4 (%identity and alignment_length, respectively). It is worthwhile (and faster) when one needs to test different thresholds.

Bring your issues on!