# Making Request to Database

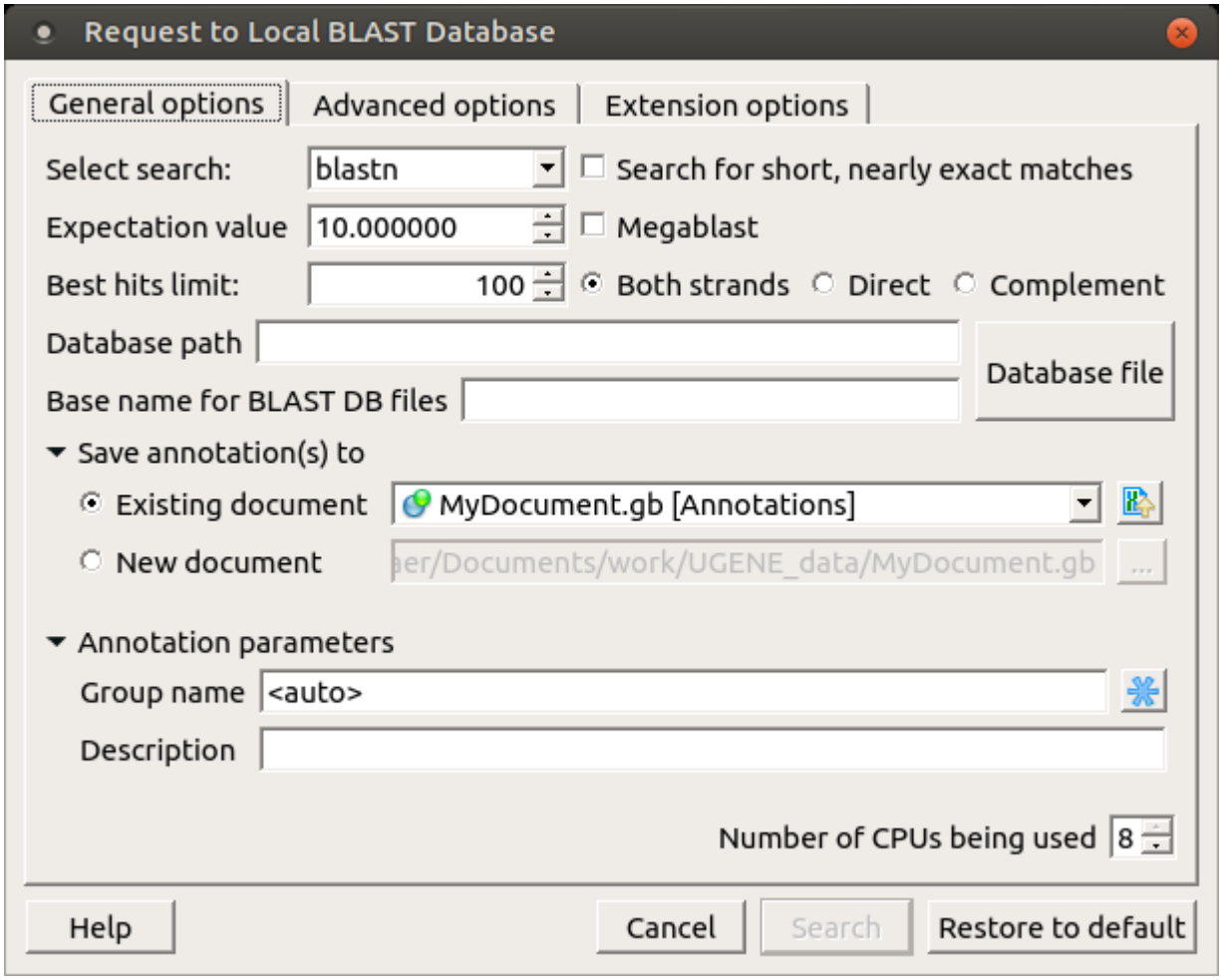
To make a request to a local BLAST database do the following:

- If you're using *BLAST* open *Tools  BLAST  BLAST Search*.
- If you're using *BLAST+* open Open *Tools  BLAST  BLAST+ Search*.

If there is a sequence opened you can also initiate the request to a local BLAST database from the *Sequence View*:

- If you're using *BLAST* select the *Analyze  Query with BLAST* item in the context menu or in the *Actions* main menu.
- If you're using *BLAST+* select the *Analyze  Query with BLAST+* item in the context menu or in the *Actions* main menu.

The *Request to local BLAST database* dialog will appear:




The following general options are available:

*Select search* - here you should select the tool you would like to use. If the query sequence is a nucleotide sequence then *blastn*, *blastx* and *tblastx* it ems are available. For a protein sequence the items are *blastp* and *tblastn*.

*Expectation value* - this option specifies the statistical significance threshold for reporting matches against database sequences. Lower expect thresholds are more stringent, leading to fewer chance matches being reported.

*Culling limit* - the maximum number of hits that will be shown (not equal to number of annotations). The maximum availablle number is 5000.

*Search for short, nearly exact matches* - automatically adjusts the word size and other parameters to improve results for short queries.

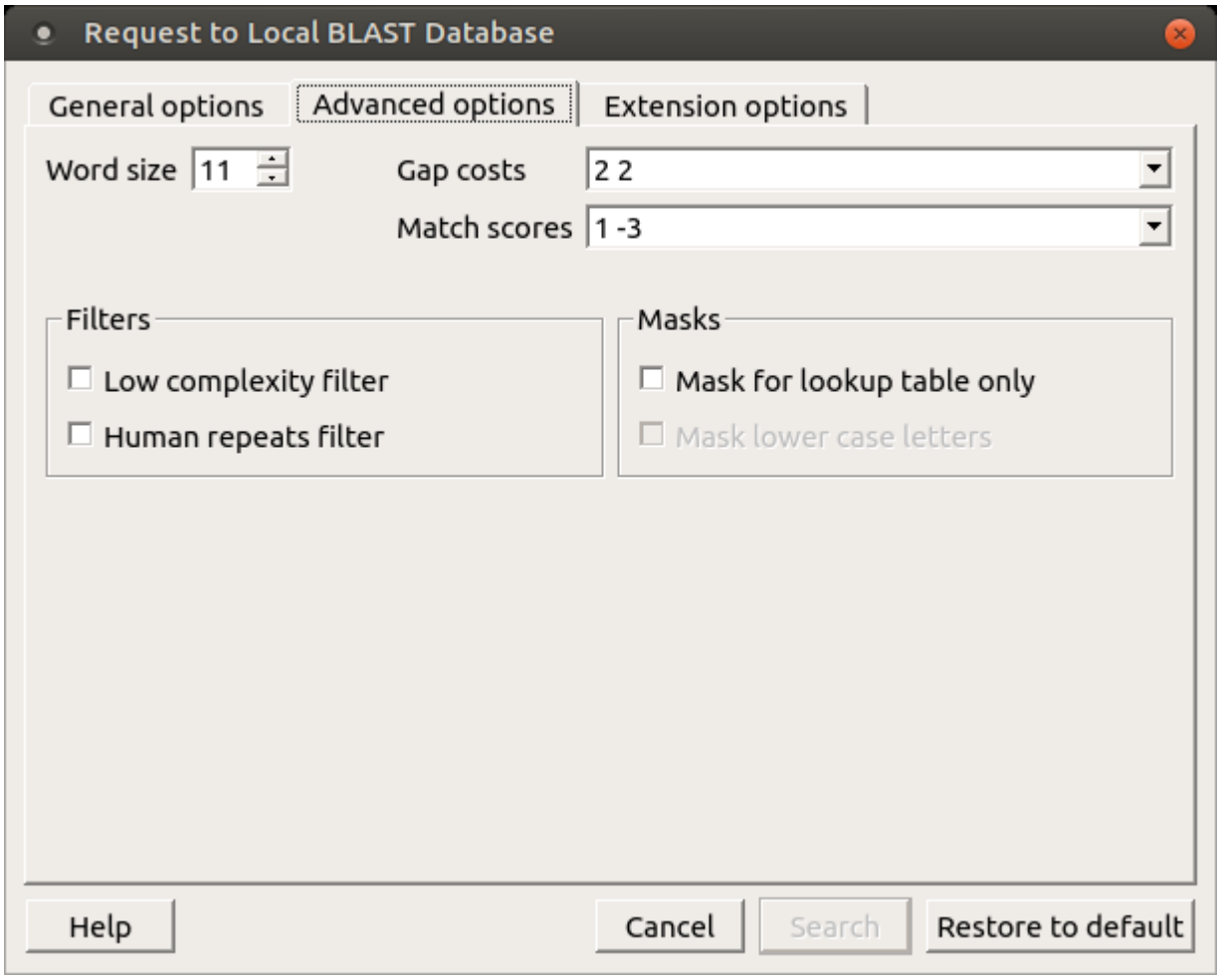
*Megablast* - select this option to compare query with closely related sequences. It works best if the target percent identity is 95% or more, but it is very fast.

*Database path* - path to the database files.

*Base name for BLAST DB files* - base name for the BLAST database files.

You can see the description of the annotation saving parameters *here*.

The following advanced parameters are available:




*Word size* - the size of the subsequence parameter for the initiated search.

*Gap costs* - costs to create and extend a gap in an alignment. Increasing the Gap costs will result in alignments which decrease the number of Gaps introduced.

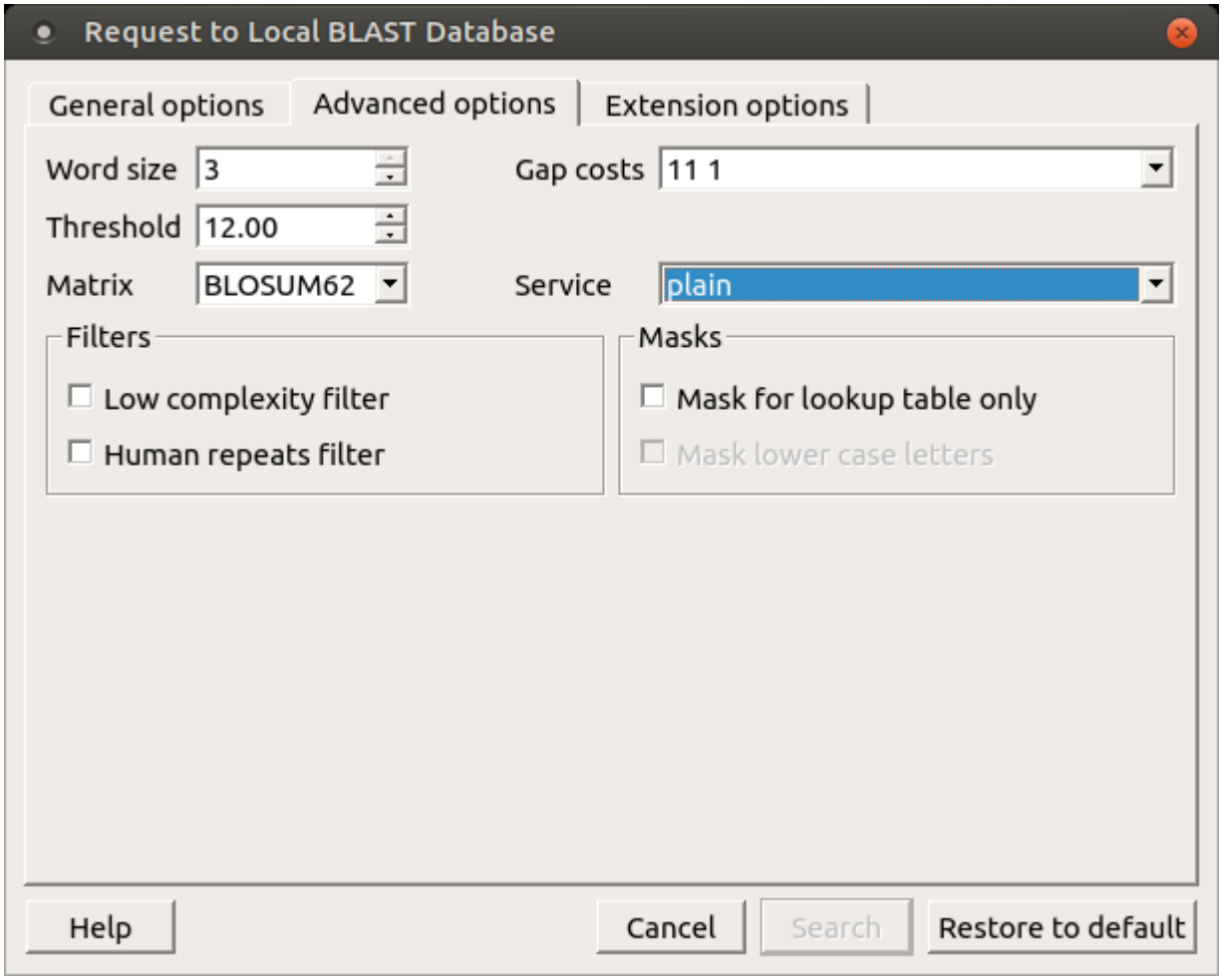
*Match scores* - reward and penalty for matching and mismatching bases.

*Filters* - filters for regions of low compositional complexity and repeat elements of the human's genome.

*Masks for lookup table only* — this option masks only for purposes of constructing the lookup table used by BLAST so that no hits are found based upon low-complexity sequence or repeats (if repeat filter is checked).

*Mask lower case letters* — with this option selected you can cut and paste a FASTA sequence in upper case characters and denote areas you would like filtered with lower case.

The view of the *Advanced options* tab depends on the selected search. For the *blastn* search it looks like on the picture above. When the *blastx* search is selected in the general options, the view of the *Advanced options* tab is the following:




As you can see there is no *Match scores* option, but there are *Threshold*, *Matrix, Composition-based statistics* and *Service* options.

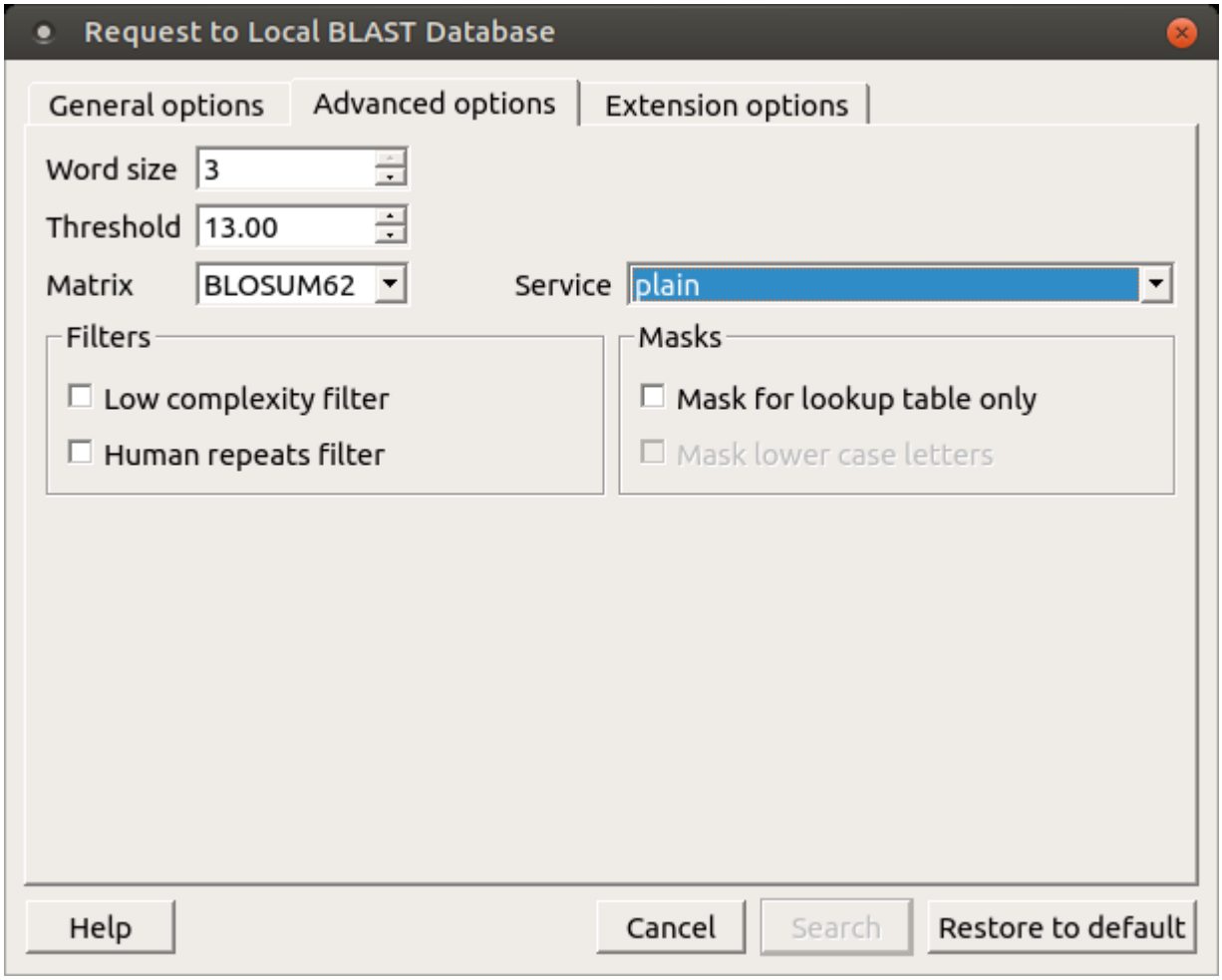
*Threshold* - threshold for extending hits.

*Matrix* — key element in evaluating the quality of a pair-wise sequence alignment is the “substitution matrix”, which assigns a score for aligning any possible pair of residues.

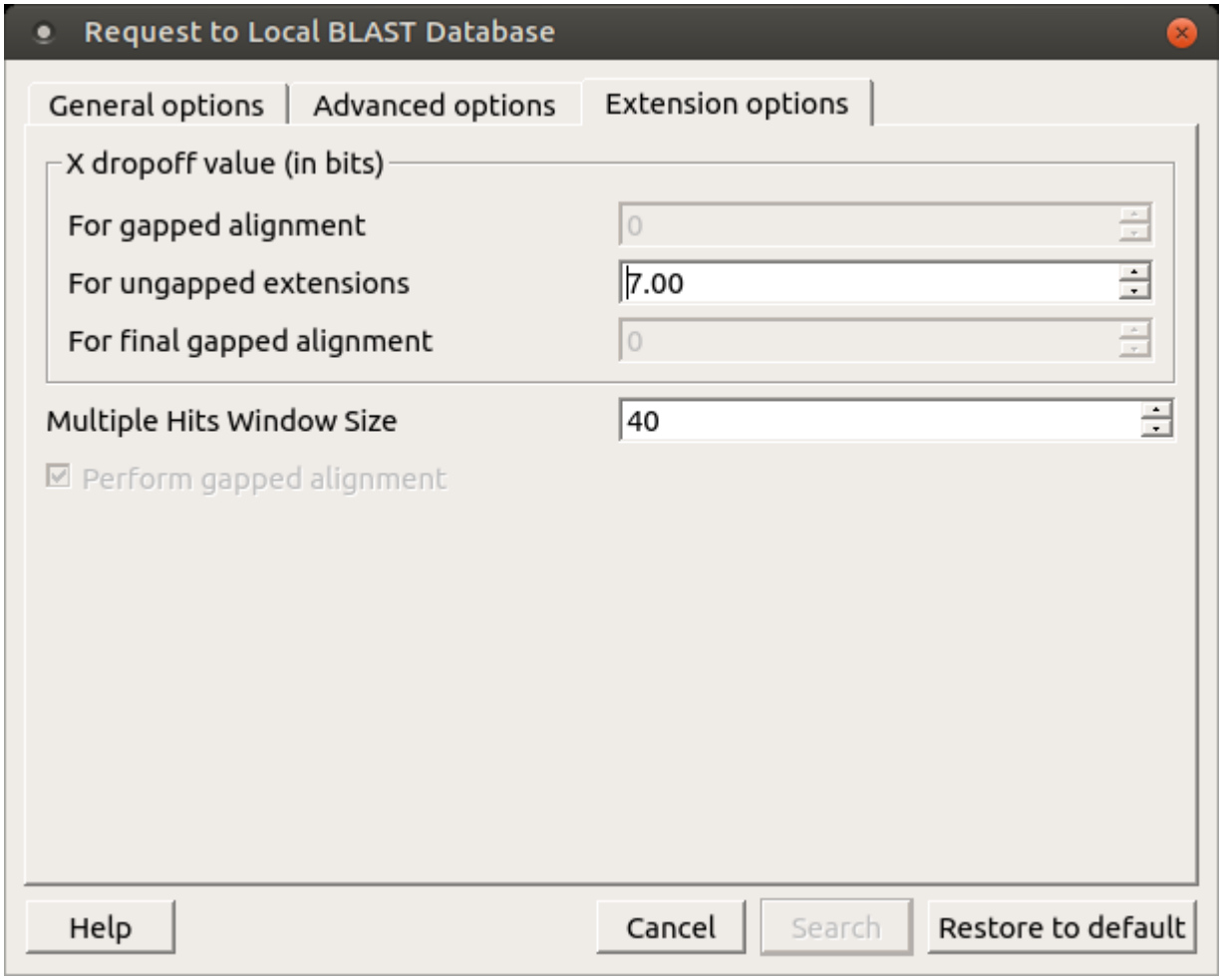
*Service* — blastp service which needs to be performed: plain, psi or phi.

*Composition-based statistics* - composition-based statistics.

When the *tblastx* search is selected in the general options, the view of the *Advanced options* tab is the following:




The following extension options are available:




*For gapped alignment* - X dropoff value (in bits) for gapped alignment.

*For ungapped alignment* - X dropoff value (in bits) for ungapped alignment.

*For final gapped alignment* - X dropoff value (in bits) for final gapped alignment.

*Multiple hits window size* - multiple hits window size.

*Perform gapped alignment* - performs gapped alignment.