

C-Scanner - Operation Manual



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What is C-Scanner?

The C-Scanner allows you to rank the selected epitopes (or any other peptide sequences) according to their complexity.

When selecting epitope sequences using various software (EpiQuest-B, A or other platforms) you would like to end up with a *unique* epitope – whether you plan developing a specific antibody or using this peptide epitope as an antigen in antibody assays.

The C-Scanner allows you to scan a group of peptide sequences and determine the most unique ones. Uniqueness of epitopes greatly define the specificity of an antibody prepared against it i.e. the sequences with low complexity will likely be present in proteins other than the target one.

The software also allows you to define a shorter, more specific, sequence within the long epitope sequences, and thus ensure the specificity of epitope used in your antibody or assay development project.

Species specificity

The matrix C1.3 is based on the complexity of proteins in *higher mammals*. Sequences from i.e. lower invertebrates, fungi, etc. may receive a lower overall score in comparison to mammals, but still, the scanning allows you to establish the most unique sequences. Whether making an antibody or looking for epitopes recognised by natural antibodies, we aim to look at the immune response in higher mammals, so the uniqueness of the tested epitopes should be defined for these types of animals.

Entering the Data

Project Name/Code

Clear

Browse

Code	Sequence
X292	GVITYSILNQEPKEPT
X573	GTLVLNLLDVNDNGPFLFPQQESFCQKDPG
X501	ATYTAQDPDKEQNQ
X119	REGHRHRQDLFSGKHSHHPK
X423	TDTGNIGLLKTVKGLDYE
X396	EDIEGTDAWNA

CADH1 XENLA B-EPITOPES

Load demo

To test the program, select Demo sequences and click **Load demo**. In the window for sequences the two columns will appear: **Code** and **Sequence**.

To empty the window, click **Clear**.

To load your own data, it should be presented in **XLS** or **XSL** format (MS-**Excel**) and to contain only *one dataset* with data per file. The data should be presented as two columns. The first should be named and contain **Code** (just your number or other identifier for the peptide; we do not recommend going above 15 signs, letters, numbers, and dash signs allowed) and **Sequence** (sequences of the peptides to be tested in a single letter code; we do not recommend going above 100 amino acids each.).

You may **Browse** for the location of the file and click on it to load.

Settings for Complexity analysis

Settings for Analysis

Matrix: C1.3

Frame size: 6 ∈ [3, 15]

Peptide size: 9 ∈ [6, 20]

Threshold: 0 ∈ [-4, 4]

Sort by: CI BEST

Default

Currently only one **Matrix** C1.3 – is available.

You can adjust your cut-off level for weak/functionally negative epitopes by changing the **Threshold** (positive numbers to decrease the sensitivity, negative – to increase). We do not recommend going beyond (-2) or (+2).

Frame size defines the size of the context analysed for every position in the sequence, we recommend keeping it at 6 (the default value).

Peptide size defines the size of epitope you are looking for in your peptides. If the tested sequences are of different length (i.e. your data set includes 10-mers, 20-mers, other), we recommend keeping it at 9, since very few epitopes and practically none of the specific ones are shorter than 9 amino acids. In case you wish to compare a set of peptides of different length and to select the best of them, set the size of the shortest one as the reported peptide size.

Sort by. You can select the data to be reported in several ways:

Start: the results will be sorted according to the order of the analysed peptides in your data file. If you analyse 19-20 mers and have defined the *Peptide size* as 9, your results will contain multiple 9-mers for every original peptide with AGI (antigenicity) defined for every 9-mer. In Results the data will be sorted according to the order of the peptide in the original sequence.

Report for "Analysis of epitopes complexity":

Code	Sequence	Start	End	Peptide	CI	CPB
X292	GVITYSILNQEPKEPT	1	9	GVITYSILN	90	10
X292	GVITYSILNQEPKEPT	2	10	VITYSILNQ	57	6
X292	GVITYSILNQEPKEPT	3	11	ITYSILNQE	21	2
X292	GVITYSILNQEPKEPT	4	12	TYSILNQEP	-12	-1
X292	GVITYSILNQEPKEPT	5	13	YSILNQEPK	-37	-4
X292	GVITYSILNQEPKEPT	6	14	SILNQEPKE	-55	-6
X292	GVITYSILNQEPKEPT	7	15	ILNQEPKEP	-63	-7
X292	GVITYSILNQEPKEPT	8	16	I NOPFKFPT	-63	-7

CI: all results will be sorted according to the order of the analysed peptides in the data file, for every analysed sequence they will be presented from the best to the worst.

CI ALL: the results will be sorted according to the AGI values of the detected peptides of the defined peptide length (say, 9) irrespective of their origin from different original sequences, from the best to the worst.

Report for "Analysis of epitopes complexity":

Code	Sequence	Start	End	Peptide	CI	CPB
X669	NVTKLHITICQ	1	9	NVTKLHITI	162	18
X669	NVTKLHITICQ	2	10	VTKLHITIC	155	17
X852	ALNDWGPRFTKLADMYGGDED	1	9	ALNDWGPRF	146	16
X852	ALNDWGPRFTKLADMYGGDED	2	10	LNDWGPRFT	139	15
X669	NVTKLHITICQ	3	11	TKLHITICQ	137	15
X852	ALNDWGPRFTKLADMYGGDED	3	11	NDWGPRFTK	135	15
X852	ALNDWGPRFTKLADMYGGDED	4	12	DWGPRFTKL	125	13
X852	ALNDWGPRFTKLADMYGGDED	5	13	WGPRFTKIA	102	11

CI BEST: the data will be presented by *only* the best peptide form every sequence in the order from the highest to the lowest (here only 1 9-mer is shown for the original 4 peptides of various lengths).

Report for "Analysis of epitopes complexity":

Code	Sequence	Start	End	Peptide	CI	CPB
X669	NVTKLHITICQ	1	9	NVTKLHITI	162	18
X852	ALNDWGPRFTKLADMYGGDED	1	9	ALNDWGPRF	146	16
X292	GVITYSILNQEPKEPT	1	9	GVITYSILN	90	10
X423	TDTGNIGLLKTVKGLDYE	8	16	LLKTVKGLD	82	9
X634	AIVTGQSILELRP	1	9	AIVTGQSIL	60	6
X158	SENEKGPFPKRI	4	12	EKGPFPKRI	59	6
X119	REGHRHRQDLFSGKHSHHPK	3	11	GHRHQDLF	57	6
X396	FDTFGTDAWNA	1	9	FDTFGTDAW	57	6

Viewing and saving the Results

You may **View** all results in new window or **Save** them for your records as HTML file. You can always import such file into your spreadsheet program or simply copy the selected areas of interest in other file formats.

Report: Relative complexity of epitopes (C-Scanner)

Date & Time: 05.11.2020 19:08:51

Project name:

Program: EpiQuest C-Scanner v1.0.0.1

Matrix: C1.3

Peptide size: 9

Frame size: 6

Threshold: 0

Sorted by: CI BEST

Code	Sequence	Start	End	Peptide	CI	CPB	
X669	NVTKLHITICQ		1	9	NVTKLHITI	162	18
X852	ALNDWGPRFTKLADMYGGDED		1	9	ALNDWGPRF	146	16
X292	GVITYSILNQEPKEPT		1	9	GVITYSILN	90	10
X423	TDTGNIGLLKTVKGLDYE		8	16	LLKTVKGLD	82	9
X634	AIVTGQSILELRP		1	9	AIVTGQSIL	60	6
X158	SENEKGPFPKRI		4	12	EKGPFPKRI	59	6
X119	REGHRHRQDLFSGKHSHHPK		3	11	GHRHRQDLF	57	6
X396	EDIEGTDANNA		1	9	EDIEGTDAN	57	6
X786	PQYRPRPANPDEIGNFIDENLN		10	18	PDEIGNFID	50	5
X753	GEEDQDFDLSQLHRGLDARP		4	12	DQDFDLSQL	43	4
X85	YIKNPAKMKD		1	9	YIKNPAKMK	37	4
X573	GTLVLNLLDVNDNGPFLEPQQESFCQKDPG		21	29	QESFCQKDP	20	2
X360	VMDTNDNPVFD		4	12	TNDNPVFD	15	1
X242	IIIKVQDQN		1	9	IIIKVQDQN	-13	-1
X501	ATYTAQDPDKEQNQ		1	9	ATYTAQDPD	-15	-1
X726	KKVVKEPLL		1	9	KKVVKEPLL	-21	-2
X834	ASLSSLNNSPNSDLQDY		9	17	PNSLDQDY	-25	-2

Demo Sequences

For Demo, we supply several files with either tested selections of epitopes or selected sets of sequences for training.