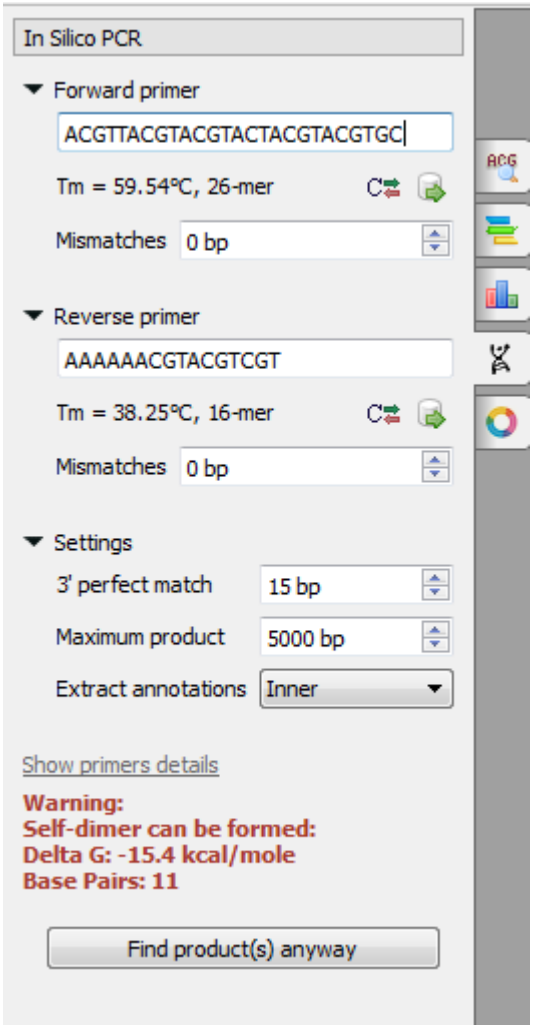
In Silico PCR

In Silico PCR Overview

In silico PCR is used to calculate theoretical polymerase chain reaction (PCR) results using a given set of primers (probes) to amplify DNA sequences.

UGENE provides the In silico PCR feature only for nucleic sequences. To use it in UGENE open a DNA sequence and go to the *In silico PCR* tab of the Options Panel:



There are the following parameters:

Forward primer - forward primer.

Reverse primer - on the opposite strand from the forward primer.

Mismatches - mismatches limit.

3' perfect match - specify the number of nucleotides at the 3' end that must not have mismatches.

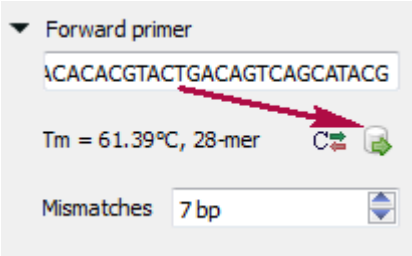
Maximum product - maximum size of the amplified sequence.

Extract annotations - specify the type of extracted annotations: *Inner, All intersected* or *None*.

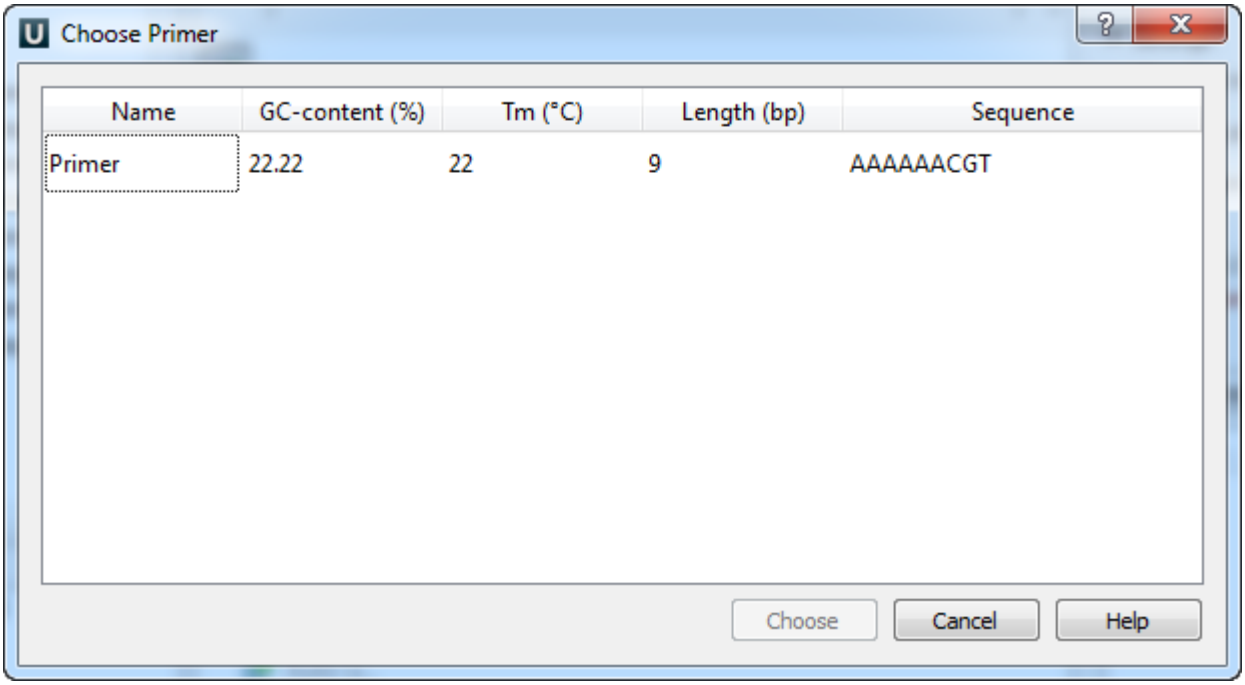
- Value *Inner* is selected by default. When this value is selected, the extracted PCR product contains annotations from the original sequence, located within the extracted region.
- Value *All intersected* specifies that all annotations of the original sequence that intersect the extracted region must be extracted as well.
- Value *None* specifies that annotations from the original sequence must not be extracted.

Choosing primers

Type two primers for running In Silico PCR. If the primers pair is invalid for running the PCR process then the warning is shown. Also, primers for the running In silico PCR can be chosen from a primer library. Click the following button to choose a primer from the primers library:

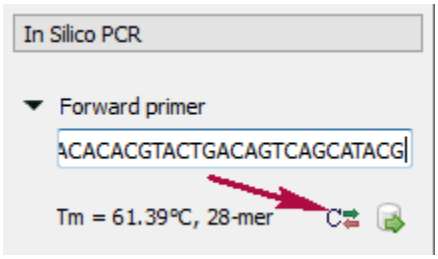


The following dialog will appear:



The table consists of the following columns: name, GC-content (%), Tm, Length (bp) and sequence. Select primer in the table and click the *Choose* button.

Click the *Reverse-complement* button for making a primer sequence reverse-complement:



Click *Show primers details* for seeing statistic details about primers.

When you run the process, the predicted PCR products appear in the products table.

Products table

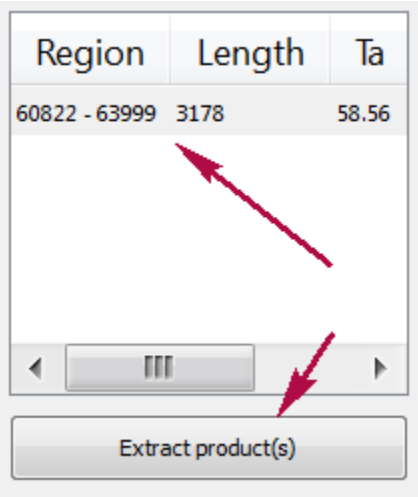
There are three columns in the table:

- region of product in the sequence

- product length
- preferred annealing temperature

Click the product for navigating to its region in the sequence.

Click the *Extract product(s)* button for exporting a product(s) in a file or use double click for that.



- Primers Details
- Primer Library