## BV-BRC Test Report

## A12. Service - Primer Design

| Item to test | Primer Design Service using bacterial and viral genes |
| :--- | :--- |
| URL | $\underline{\underline{\text { https://www.bv-brc.org/app/PrimerDesign }}}$ |
| Prerequisites | Bacterial Fasta contig files in Workspace |
| References | $\underline{\text { https://www.bv-brc.org/docs/quick references/services/primer design service.html }}$ https://www.bv-brc.org/docs/tutorial/primer design/primer design.html |
| Tester(s) | Rebecca Wattam, Maulik Shukla |
| Test date | 08-May-2022 (follow-up from original test) |
| Test result | Passed |

## Overview

- Test the Primer Design Service using exemplar bacterial and viral genes.
- Test input options, i.e., using FASTA sequence as query or a FASTA sequence file from the workspace.
- For each job submitted, verify successful completion of the job, presence of output files, and quality of the results.


## Test Data

| Dataset | Rational | Input Format | Input |
| :--- | :--- | :--- | :--- |
| SARSCoV2 Spike <br> protein | Viral gene of <br> interest | FASTA <br> sequence | sarscov2_spike_protein_na.fasta |
| MTB ahpD gene | Bacterial gene <br> of interest | FASTA <br> sequence | mtb_ahpd_protein_na.fasta |

- All test datasets and corresponding job results are available in the following public workspace: https://www.bv-brc.org/workspace/BVBRC@patricbrc.org/BVBRC\ Tests/Primer\ Design


## Test Results

- All jobs completed successfully, without any errors.
- All jobs resulted in expected output files in corresponding job output directory, providing primer report in HTML format.
- The primer report was informative and provided a concise summary of the primers and their sequences.
- All test datasets and corresponding job results are available in the following public workspace: https://www.bv-brc.org/workspace/BVBRC@patricbrc.org/BVBRC\ Tests/Primer\ Design
- Below are a series of screenshots showing successful completion of the jobs, availability of the result files in the workspace, and the primer report.


Sequence ID: figl83332.12.peg.2721|Rv2429|VBIMycTub87468_2721|
Sequence length: 534
Note: sequence positions and pair indexes are 1-based here, but 0-based in Primer3 output.

| Pair\# | Region | Sequence ( $5^{\prime} \rightarrow 3$ 3') | Start | End | Len | $\mathrm{T}_{\mathrm{m}}$ | GC\% | Any Compl. | End Compl. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | Forward primer | aacctgagctcaatcacccg | 58 | 77 | 20 | 60.04 | 55 | 18.85 | 3.79 |
|  | Reverse primer | tcagcgccaatgtcagctaa | 173 | 154 | 20 | 60.04 | 50 | 2.29 | 0.00 |
|  | Internal oligo | gcagcgtgctcgaccaggaa | 80 | 99 | 20 | 59.70 | 65 | 10.15 | 0.00 |
|  | Product/Primer pair |  | 58 | 173 | 116 |  |  | 0.00 | 0.00 |
| 2 | Forward primer | aacctgagctca | 58 | 77 | 20 | 60.04 | 55 | 18.85 | 3.79 |
|  | Reverse primer | ttcagcgccaatgtcagcta | 174 | 155 | 20 | 60.04 | 50 | 2.29 | 0.00 |
|  | Internal oligo | gcagcgtgctcgaccaggaa | 80 | 99 | 20 | 59.70 | 65 | 10.15 | 0.00 |
|  | Product/Primer pair |  | 58 | 174 | 117 |  |  | 0.00 | 0.00 |
| 3 | Forward primer | aacctgagctcaatcacccg | 58 | 77 | 20 | 60.04 | 55 | 18.85 | 3.79 |
|  | Reverse primer | ggccacggtagaacacgtta | 262 | 243 | 20 | 60.04 | 55 | 0.00 | 0.00 |
|  | Internal oligo | gcagcgtgctcgaccaggaa | 80 | 99 | 20 | 59.70 | 65 | 10.15 | 0.00 |
|  | Product/Primer pair |  | 58 | 262 | 205 |  |  | 0.00 | 0.00 |
| 4 | Forward primer | tagctgacattggcgctgaa | 155 | 174 | 20 | 60.04 | 50 | 15.94 | 0.20 |
|  | Reverse primer | ggccacggtagaacacgtta | 262 | 243 | 20 | 60.04 | 55 | 0.00 | 0.00 |
|  | Internal oligo | gagccgcggccatcatggg | 218 | 236 | 19 | 60.54 | 73 | 40.62 | 3.56 |
|  | Product/Primer pair |  | 155 | 262 | 108 |  |  | 0.00 | 0.00 |
| 5 | Forward primer | ttagctgacattggcgctga | 154 | 173 | 20 | 60.04 | 50 | 15.94 | 7.63 |
|  | Reverse primer | ggccacggtagaacacgtta | 262 | 243 | 20 | 60.04 | 55 | 0.00 | 0.00 |
|  | Internal oligo | gagccgcggccatcatggg | 218 | 236 | 19 | 60.54 | 73 | 40.62 | 3.56 |
|  | Product/Primer pair |  | 154 | 262 | 109 |  |  | 0.00 | 0.00 |

Sequence ID: figl2697049.107626.CDS.3IGU280_gp02|
Sequence length: 3822
Note: sequence positions and pair indexes are 1-based here, but 0-based in Primer3 output.

| Pair\# | Region | Sequence ( $5^{\prime} \rightarrow 3^{\prime}$ ) | Start | End | Len | $\mathrm{T}_{\mathrm{m}}$ | GC\% | Any Compl. | End Compl. |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | Forward primer | tgcagatgctggettcatca | 2484 | 2503 | 20 | 60.03 | 50 | 17.30 | 5.36 |
|  | Reverse primer | tgtacccgctaacagtgcag | 2643 | 2624 | 20 | 60.04 | 55 | 0.00 | 0.00 |
|  | Internal oligo | acggccttactgtttgecacct | 2567 | 2589 | 23 | 59.00 | 52 | 0.00 | 0.00 |
|  | Product/Primer pair |  | 2484 | 2643 | 160 |  |  | 0.00 | 0.00 |
| 2 | Forward primer | tttcaaacacgtgcaggctg | 1927 | 1946 | 20 | 59.90 | 50 | 17.22 | 10.94 |
|  | Reverse primer | ctagctacactacgtgeccg | 2066 | 2047 | 20 | 59.97 | 60 | 0.00 | 0.00 |
|  | Internal oligo | tgtgacatacccattggtgcaggt | 1984 | 2007 | 24 | 58.31 | 50 | 0.00 | 0.00 |
|  | Product/Primer pair |  | 1927 | 2066 | 140 |  |  | 6.99 | 2.17 |
| 3 | Forward primer | tttcaaacacgtgcaggctg | 1927 | 1946 | 20 | 59.90 | 50 | 17.22 | 10.94 |
|  | Reverse primer | cccgccgaggagaattagtc | 2050 | 2031 | 20 | 59.97 | 60 | 0.00 | 0.00 |
|  | Internal oligo | tgtgacatacccattggtgcaggt | 1984 | 2007 | 24 | 58.31 | 50 | 0.00 | 0.00 |
|  | Product/Primer pair |  | 1927 | 2050 | 124 |  |  | 0.00 | 0.00 |
| 4 | Forward primer | tgcagatgctggettcatca | 2484 | 2503 | 20 | 60.03 | 50 | 17.30 | 5.36 |
|  | Reverse primer | tctgtgagcaaaggtggcaa | 2600 | 2581 | 20 | 60.11 | 50 | 0.00 | 0.00 |
|  | Internal oligo | tgctgctagagacctcatttgtgca | 2532 | 2556 | 25 | 58.47 | 48 | 0.00 | 0.00 |
|  | Product/Primer pair |  | 2484 | 2600 | 117 |  |  | 0.00 | 0.00 |
| 5 | Forward primer | tgcagatgctggcttcatca | 2484 | 2503 | 20 | 60.03 | 50 | 17.30 | 5.36 |
|  | Reverse primer | atctgtgagcaaaggtggca | 2601 | 2582 | 20 | 59.89 | 50 | 0.00 | 0.00 |
|  | Internal oligo | tgctgctagagacctcatttgtgca | 2532 | 2556 | 25 | 58.47 | 48 | 0.00 | 0.00 |
|  | Product/Primer pair |  | 2484 | 2601 | 118 |  |  | 0.00 | 0.00 |

## References

- Primer Design Service Quick Reference Guide
- Primer Design Service Tutorial

