Hannah Itell

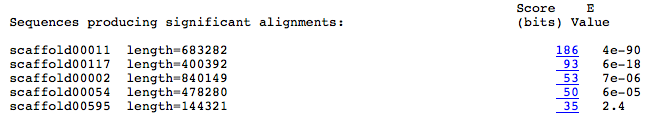
Davidson College

Enzyme: dihydrofolate reductase / thymidylate synthase (EC: 1.5.1.3)

**Introduction:**

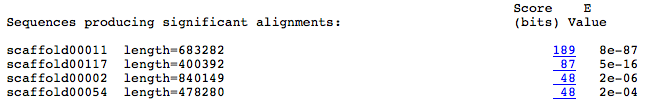
I found Vitis vinifera (wine grape) enzyme [1.5.1.3](http://www.genome.jp/dbget-bin/www_bget?vvi:100255872+vvi:100267529) (Entry: 100255872) on [Kegg Pathway](http://www.genome.jp/kegg-bin/show_pathway?org_name=vvi&mapno=00790&mapscale=&show_description=hide) for Vitis vinifera. I blasted grape 1.5.1.3 protein sequence (580 aa) against V. corymbosum 454-Scaffolds using tblastn:

**Fig. 1** Hits from tblastn of grape EC 1.5.1.3 (query) and V. corymbosum scaffolds (subject).



I also found Arabidopsis thaliana enzyme [1.5.1.3](http://www.genome.jp/dbget-bin/www_bget?ath:AT2G16370+ath:AT4G34570) (Entry: AT2G16370) on [Kegg Pathway](http://www.genome.jp/kegg-bin/show_pathway?org_name=ath&mapno=00790&mapscale=&show_description=hide) for Arabidopsis thaliana, to see if a tblastn with Arabidopsis would be any different than with Vitis vinifera. I blasted Arabidopsis 1.5.1.3 protein sequence (519 aa) against V. corymbosum 454-Scaffolds using tblastn:

**Fig. 2** Hits from tblastn of Arabidopsis EC 1.5.1.3 (query) and V. corymbosum scaffolds (subject).



When comparing the two sets of hits, I found that the same scaffolds had hits on both blasts with similar E-values. The grape blast had smaller E-values, so I used the grape results.

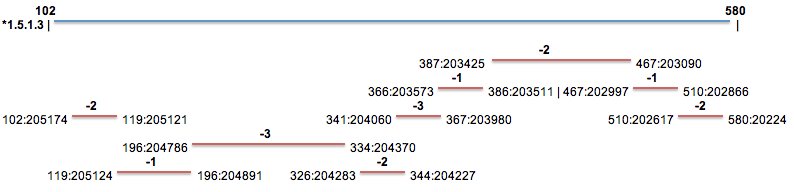
**tBlastn Results**

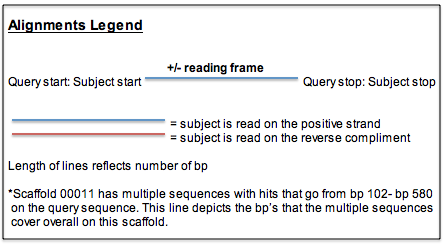
Subject**:** V. corymbosum 454-Scaffolds

Query**:** Vitis vinifera (wing grape) enzyme [1.5.1.3](http://www.genome.jp/dbget-bin/www_bget?vvi:100255872+vvi:100267529) (580 aa)

**Alignments**

**Scaffold 00011 (E = 4e-90)**



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**Scaffold 00117 (E = 6e-18)**

**+1**

412:332617505:332916

**Scaffold 00002 (E = 7e-06)**

**-2**

412:314131 475:313961

**Scaffold 00054 (E = 6e-05)**

**-2**

398:421461:149

**SSR Primers**

EC 1.5.1.3 found in Scaffold 00011 (query starts at base 205174 on scaffold)

1)

For Primer: CGCACGCAAGTTAATAGATCTTC

Rev Primer: TTTTCCCAGATTATTGACAGCC

Repeats (at) x 24 PCR Product = 279 bp & starts at base 299383

2)

For Primer: TGCTTATTGTGTACCCATTGGA

Rev Primer: GTGGTTTGGGTCTCTCTGTGTA

Repeats (taa) x 15 PCR Product = 184 bp & starts at base 252504

3)

For Primer: CTTCATCATCGACTTCAGCAAG

Rev Primer: CCCGAGTCTTTATGGTGCTTAT

Repeats (ga) x 14 PCR Product = 276 bp & starts at base 268832

EC 1.5.1.3 found in Scaffold 00117 (query starts at base 332617 on scaffold)

1)

For Primer: AAAACAACTGCAACACATCAGG

Rev Primer: GGGAGGTAGGTAGGTAGGTTGG

Repeats (ct) x 15 PCR Product = 194 bp & starts at base 272344

2)

For Primer: ATGAGAAGCGCGTTAAAGAAAC

Rev Primer: GCTCTTGTTGGCTTTGTACCTT

Repeats (ga) x 10 PCR Product = 278 bp & starts at base 394372

3)

For Primer: ATGCCGTTAGTTTTCGTCACTT

Rev Primer: GGTAGTCATTTTGGTGGGAAAA

Repeats (ga) x 9 PCR Product = 141 bp & starts at base 317477

EC 1.5.1.3 found in Scaffold 00002 (query starts at base 314131 on scaffold)

1)

For Primer: GTATGCTGCTGCAAACAATGAT

Rev Primer: CTTGGAGAAGTGACAGAAAGCA

Repeats (tc) x 44 PCR Product = 236 bp & starts at base 341579

2)

For Primer: TTTGGCGAACGAACTATGTCTA

Rev Primer: TCTAGTCATGTGGCTTCATGGT

Repeats (tc) x 20 PCR Product = 261 bp & starts at base 361596

3)

For Primer: CAAGTACAGGCCTATCGAAACC

Rev Primer: ATAGAGAGTGTGAGGCTCGCTT

Repeats (ga) x 16 PCR Product = 283 bp & starts at base 394191

EC 1.5.1.3 found in Scaffold 00054 (query starts at base 421 on scaffold)

1)

For Primer: AGCTCCGACAACGATAACAACT

Rev Primer: ACAGAAAGACAACCGGAGAGAG

Repeats (ct) x 16 PCR Product = 264 bp & starts at base 99432

2)

For Primer: GTGTGTGTGTGTGTGTGTGTGA

Rev Primer: GTCCCACATTACCTGGCTACAT

Repeats (ag) x 13 PCR Product = 138 bp & starts at base 60988

3)

For Primer: ACATCTAATGGTTCGGATCGTC

Rev Primer: CTGAGCCCAAAGGTTTATATGG

Repeats (ta) x 10 PCR Product = 275 bp & starts at base 84700