

uMelt HETSSM User Guide

v2.4

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Introduction to uMelt HETS

- **Functionality**
 - Predict heteroduplex/homoduplex melting transitions
 - Predict composite heterozygote from individual duplexes

Parameters and Input

- User sequence with SNP location marked with an 'x'
- Variation found in drop down box
- Laboratory conditions included :
 - Monovalent cation concentration
 - free Mg⁺⁺ concentration
 - Betaine & DMSO%
 - Temperature range
- Nearest-neighbor thermodynamic parameters are included from the SantaLucia and Hicks (2004) publication
 - <http://www.ncbi.nlm.nih.gov/pubmed/15139820>

Quick Overview

- Paste amplicon sequence into interface
 - Mark SNP location with ‘x’
- Adjust Parameters
- Push ‘Analyze’ button to run algorithm
- Download Chart Image (.png) or data file

1) Paste sequence into sequence box located under chart

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Genotype Predictions Individual Duplexes

100 80 60 40 20 0

0 20 40 60 80 100

100 80 60 40 20 0

0 20 40 60 80 100

Parameters Standard Temperature Range Disclaimer

[Mono⁺] 20 mM Betaine 0 M

free [Mg⁺⁺] 2 mM

DMSO % 0

Parameter Definitions

Sequence Variant Index 26

TCTCTGTATTTCTTACTCTAAGTGCxTATTTTCATGAGAACTGGTTTTTCATAG

Variation A/G Analyze Download Data

1b) Select variation from drop down box.

The image displays the uMelt HETS v2.4 software interface. At the top, there is a green banner with a DNA double helix and the text "uMelt HETS v2.4". Below this, there are two tabs: "Genotype Predictions" and "Individual Duplexes". The main area contains two identical empty bar charts with y-axes from 0 to 100 and x-axes from 0 to 100. Below the charts is a legend for "Genotype" with three options: G/G (black line), A/A (blue line), and A/G (red line). At the bottom, there is a "Parameters" section with input fields for [Mono⁺] (20 mM), free [Mg⁺⁺] (2 mM), DMSO % (0), and Betaine (0 M). To the right is a "Sequence" section with a text box containing "TCTCTGTATTTTCTTACTC...ATATTTTCATGAGAACTGGTTTTTCATAG" and a "Variant Index 26". Below the sequence is a "Variation" dropdown menu currently set to "A/G", an "Analyze" button, and a "Download Data" button. A large orange arrow points to the "A/G" dropdown menu.

2) Adjust parameters



3) Push Analyze button

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Genotype Predictions Individual Duplexes

100 80 60 40 20 0

0 20 40 60 80 100

Genotype — G/G — A/A — A/G

Parameters Standard Temperature Range Disclaimer

[Mono⁺] 20 mM Betaine 0 M

free [Mg⁺⁺] 2 mM

DMSO % 0

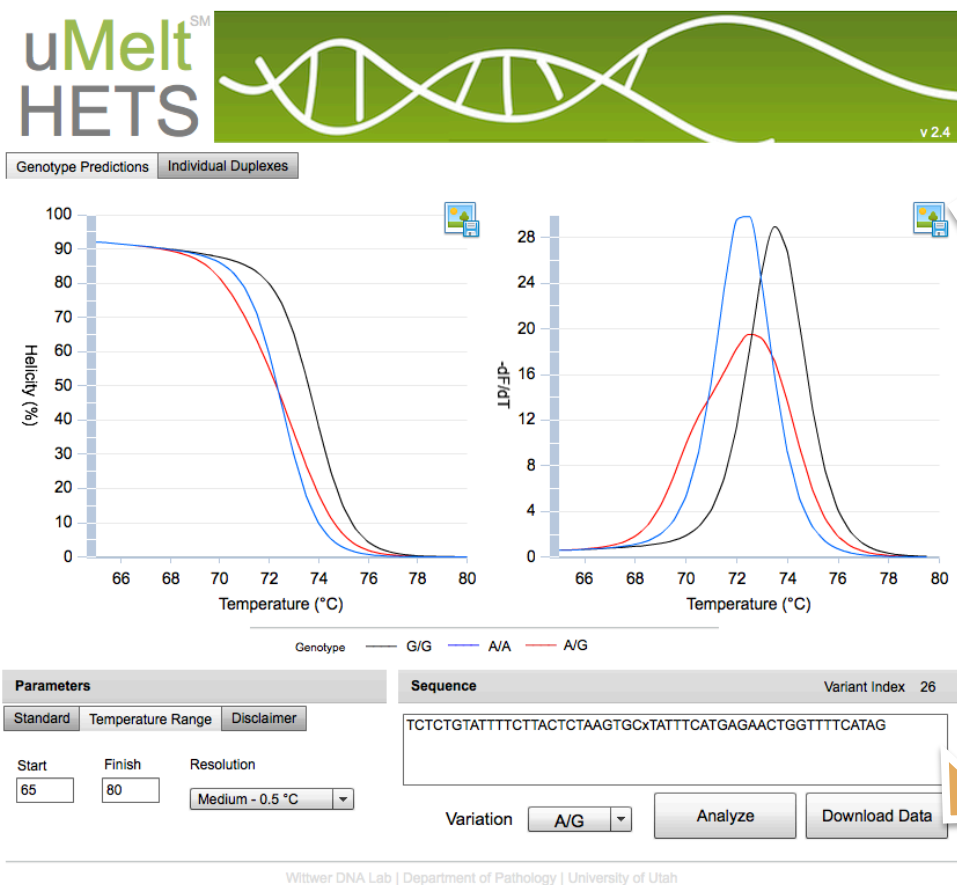
Parameter Definitions

Sequence TCTCTGTATTTTCTTACTCTAAGTGCxTATTCATCAGTCATAG

Variation A/G

Analyze Download Data

3) Result wills appear



Images can be downloaded here

Temperature range may need to be adjusted

Data may be downloaded via button push

Predictions

- A predicted melting curve is hardcoded to use the following parameters:
 - **Thermodynamics**
 - SantaLucia & Hicks (*Pubmed ID* : 15139820)
 - **Melting Algorithm**
 - uMelt (*Pubmed ID* : 21300699)
 - **Cooperativity**
 - Blossey and Carlon (*Pubmed ID* : 14754238)

Questions & Comments

- We welcome feedback, questions, and comments about all of our software.
 - Lab Contact : Zach Dwight
 - Email : zach.dwight@path.utah.edu

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