## HRMType®: a brief user's manual

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This is a brief description of how to use HRMType<sup>®</sup>, a program in STATA which allows the generation of typing schemes using a high resolution melt platform to interrogate amplified fragments.

The first section describes how to move the data from the MLST database into Stata in a way which allows Stata to use the full concatenated sequences.

- Download the concatenated sequences from the MLST database as an Access file. Concatenate the whole sequence. Stata is unable to manage string variables of longer than 244, so cut the concatenated sequence up into 244 length fragments. Use the Mid([seq],1,244) command to do this. The name of each field should be named: st seq\_1 seq\_245 seq\_489 seq\_733 seq\_977 seq\_1221 seq\_1465 seq\_1709 seq\_1953 seq\_2197 seq\_2441 seq\_2685 seq\_2929 seq\_317
- 2. Cut and paste the table with the sequence types and corresponding 244 length sequences into Stata.
- 3. The HRMType do file generates the predicted curves. The main inputs are:
  - a. Which sequence types you want to drop from the analysis.
  - b. How many regions are to be interrogated.
  - c. The start and end positions of these regions.

The do file is named "generating\_hrm\_curve\_list.do". An example is below

clear			
set mem 80m			
use "U:\infectious-diseases\HRMType\S aureus\staph	mlst i	n 244.dta",	clear
destring st, replace			

drop if length!= drop if st==

local	regions=5
local	start1=150
local	end1=300
local	start2=1450
local	end2=1600
local	start3=457
local	end3=620
local	start4=2900
local	end4=3050
local	start5=2300
local	end5=2530

After running the file, the output you will see at the bottom of the screen will be the D value for your chosen regions. Further output can be obtained such as:

. list st b\* MelT if st<21 +----+ | st b1 b2 b3 b4 b5 b6 MelT | **Comment [ST1]:** You can add or subtract which sequence types you want to exclude from the analysis.

**Comment [ST2]:** Choose how many regions to interrogate.

**Comment [ST3]:** Enter the start and end positions on the concatenated sequence of the region of interest. Note this is not where the primers start and end, but the amplified region inside of the primers.

Continue this for each of the regions of interest. To add more regions, just create further lines.

1. 2. 3. 4. 5.	1   2   3   4   5	53 51 53 53 53	24 24.5 24 24.5 23	13 12 13 12 12 12	43 45 43 44 43	66 65 66 65 66	43 43 43 44 43	202   62   202   213   174
6. 7. 8. 9. 10.	   6   7   8   9   10	52 52 52 52 52 52	23 23 23 23 23 23	12 12 13 13 12	43 44 44 43 43	66 65 65 66 65	43 43 43 43 43 44	87   91   103   100   86
11. 12. 13. 14. 15.	11   12   13   14   15	53 53 53 53 53 54	23 23 23 22 22 22	12 12 12 13 13	43 44 44 43 43	66 65 65 65 65	43 44 44 43 43	174   178   178   178   164   237
16. 17. 18. 19. 20.	16   17   18   19   20	53 51 54 51 51 54	22 23.5 22 24.5 23	13 12 13 12 12 12	43 44 43 44 43	65 65 65 62 65	43 42 43 43 43	164   34   237   55   240

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