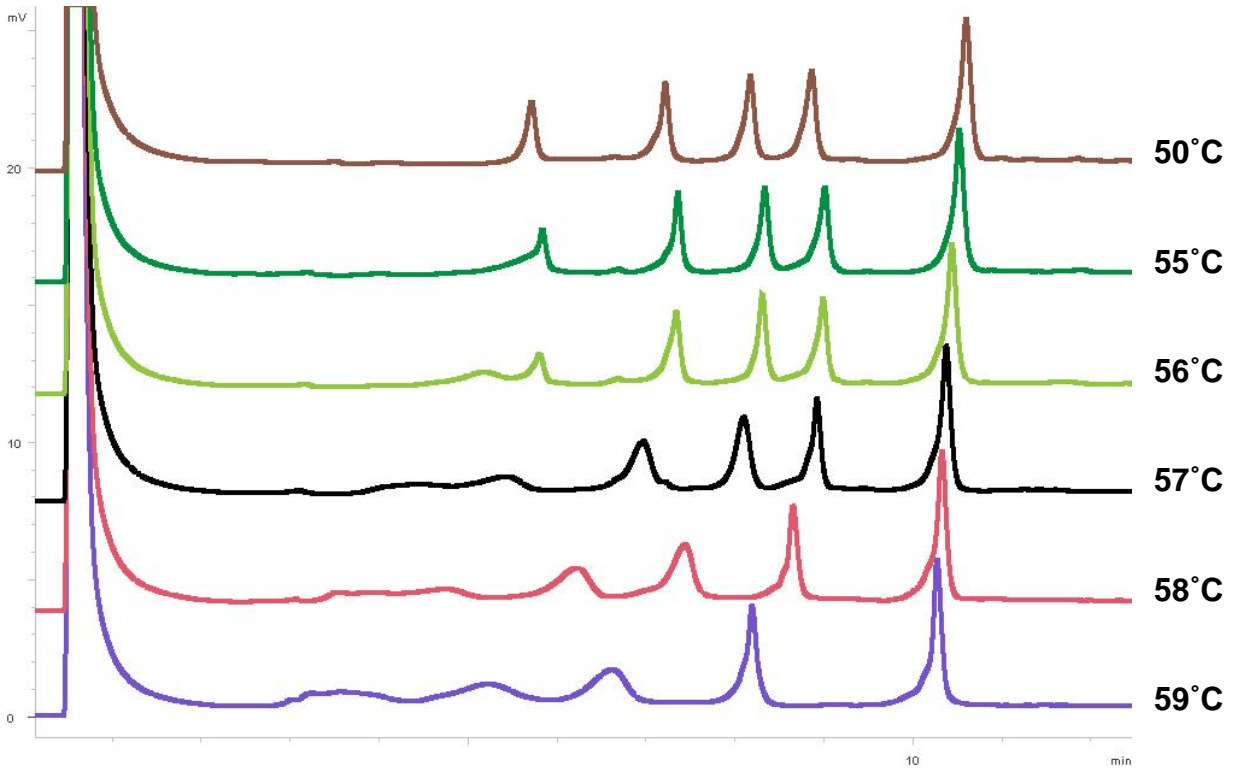


G8363A Mutation

Figure 1 DHPLC profiles of 40% G8363A mutation at different temperatures.

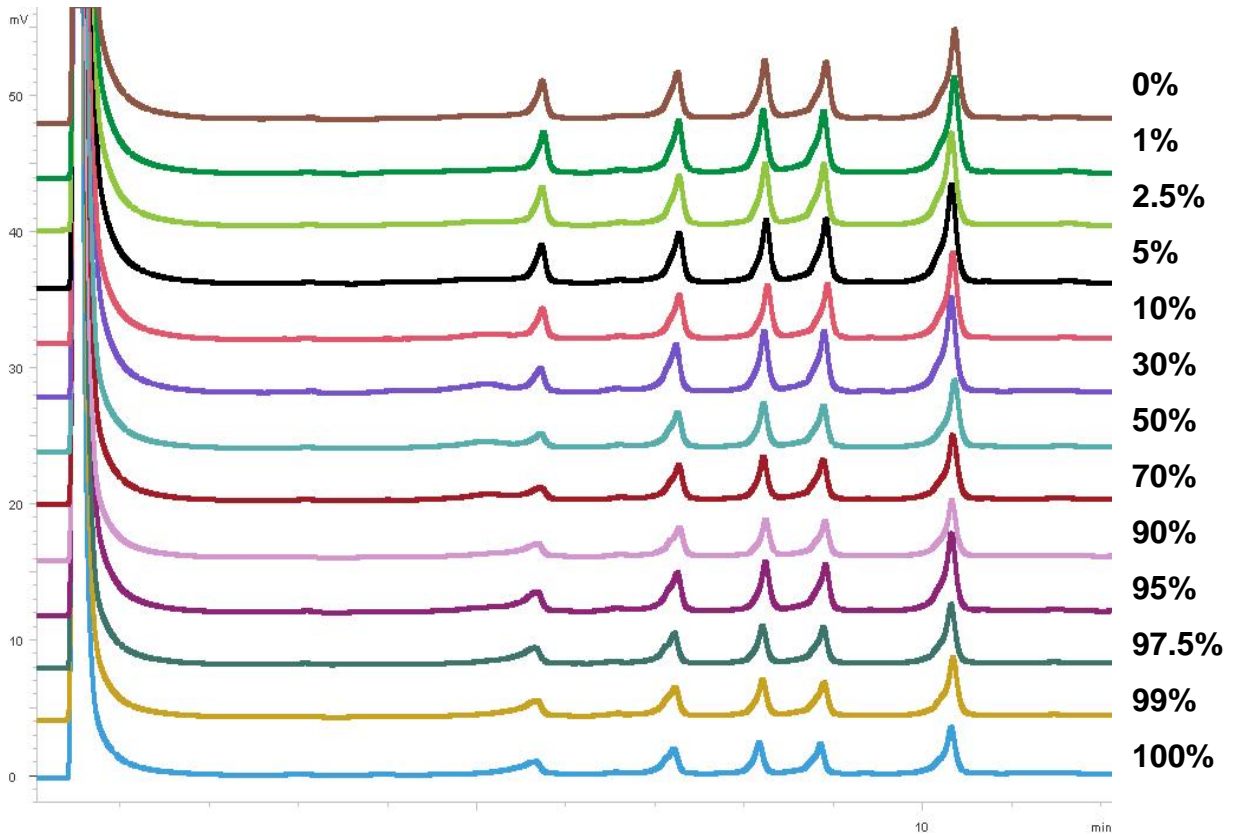


DHPLC Gradient

Step	Time	%A	%B
Loading	0	60	40
Start Gradient	0.1	55	45
Stop Gradient	12.1	31	69
Start Clean	12.2	0	0
Stop Clean	13.2	0	0
Start Equilibrate	13.3	60	40
Stop Equilibrate	15.8	60	40

Temperature: 50, 55, 56, 57, 58, 59°C

Figure 2 DHPLC profiles of 0%-100% G8363A mutations at the optimized DHPLC temperature.



DHPLC Gradient

Step	Time	%A	%B
Loading	0	60	40
Start Gradient	0.1	55	45
Stop Gradient	12.1	31	69
Start Clean	12.2	0	0
Stop Clean	13.2	0	0
Start Equilibrate	13.3	60	40
Stop Equilibrate	15.8	60	40

Optimum Temperature: 56°C

PCR condition

PCR was performed in 70 μ L Optimase reaction buffer containing 150 ng total cellular DNA, 200 μ mol/L of each dNTP, 21 pmol of the forward and reverse primer, and 3.5 units Optimase DNA polymerase (Transgenomic).

The conditions for PCR were as follows: 95°C for 2 min; 35 cycles of 95°C for 30 s, 56°C for 30 s, and 72°C for 3 min; and a final extension step of 72°C for 5 min.