FIGURE 1

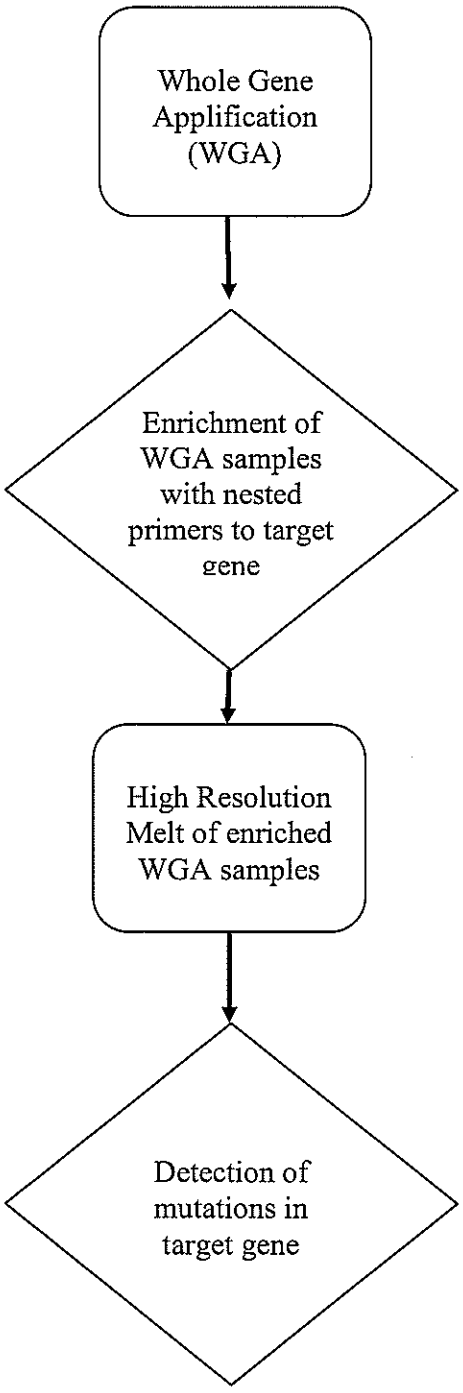

Whole Gene Applification (WGA)
Enrichment of WGA samples with nested primers to target gene
High Resolution Melt of enriched WGA samples
Detection of mutations in target gene

FIGURE 2A

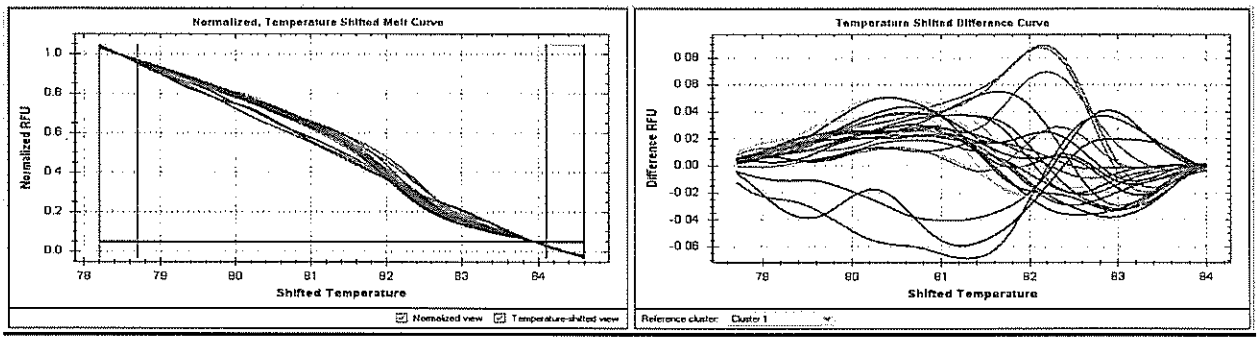


FIGURE 2B

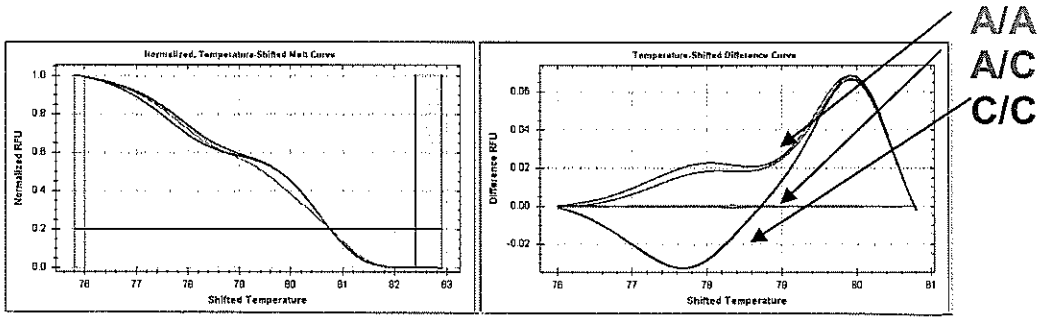


FIGURE 2C

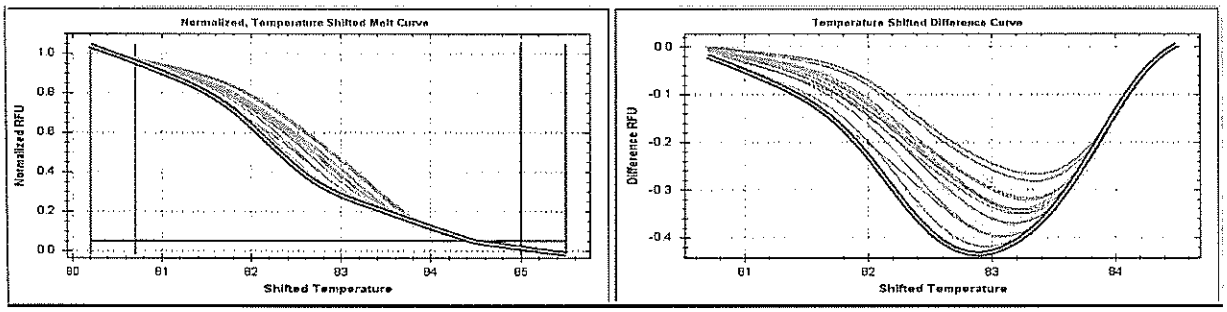


FIGURE 2D

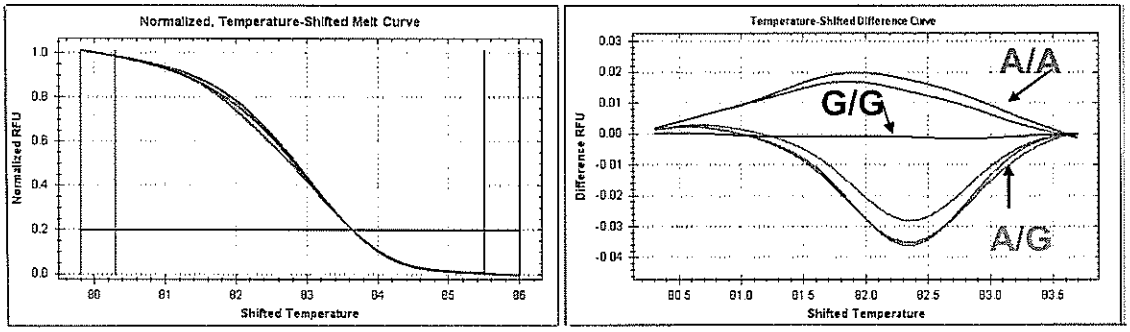
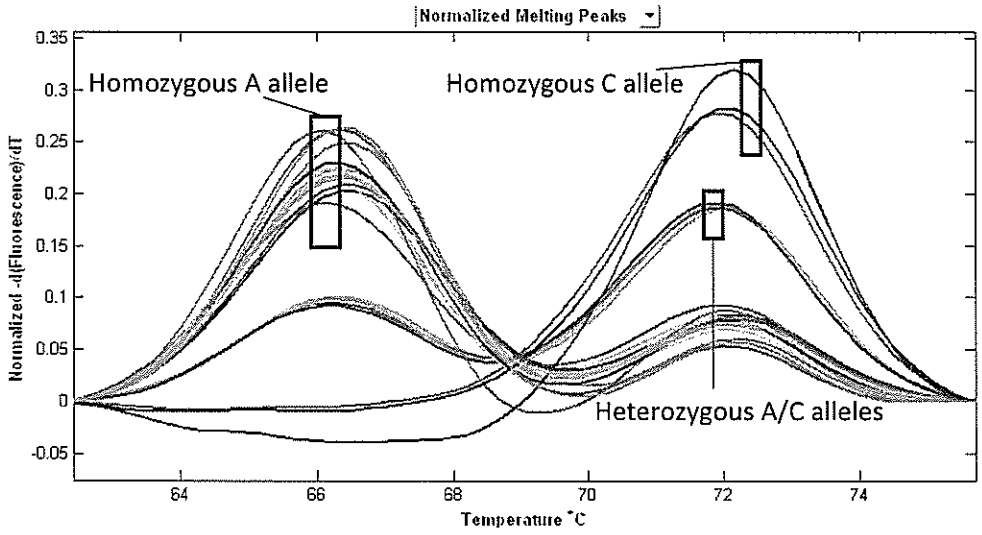
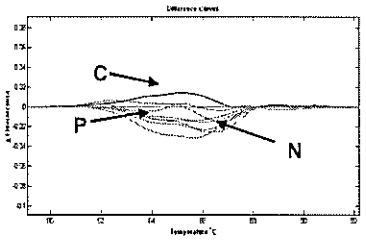


FIGURE 3

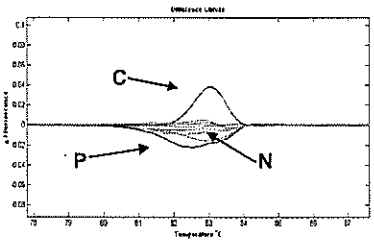

Normalized Melting Peaks
Homozygous A allele
Homozygous C allele
Heterozygous A/C alleles
Normalized -d(Fluorescence)/dT
Temperature °C
0.35
0.3
0.25
0.2
0.15
0.1
0.05
0
-0.05
64
66
68
70
72
74

A **B**

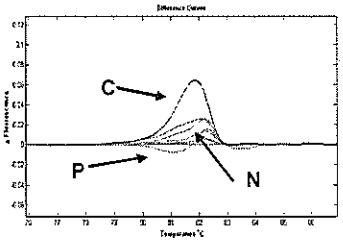
FIGURE 4



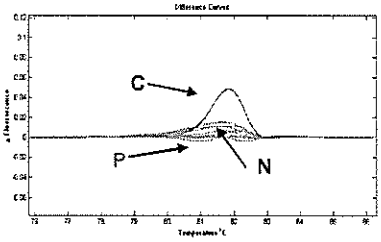
SYBR Green 1



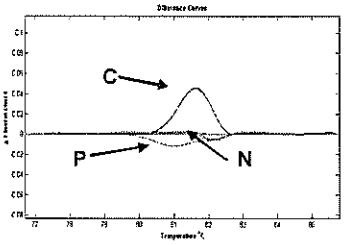
LCGreen Plus



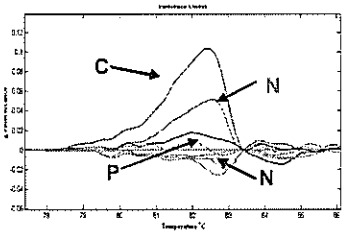
ResoLight



EvaGreen



SYTO9



BEBO

FIGURE 5

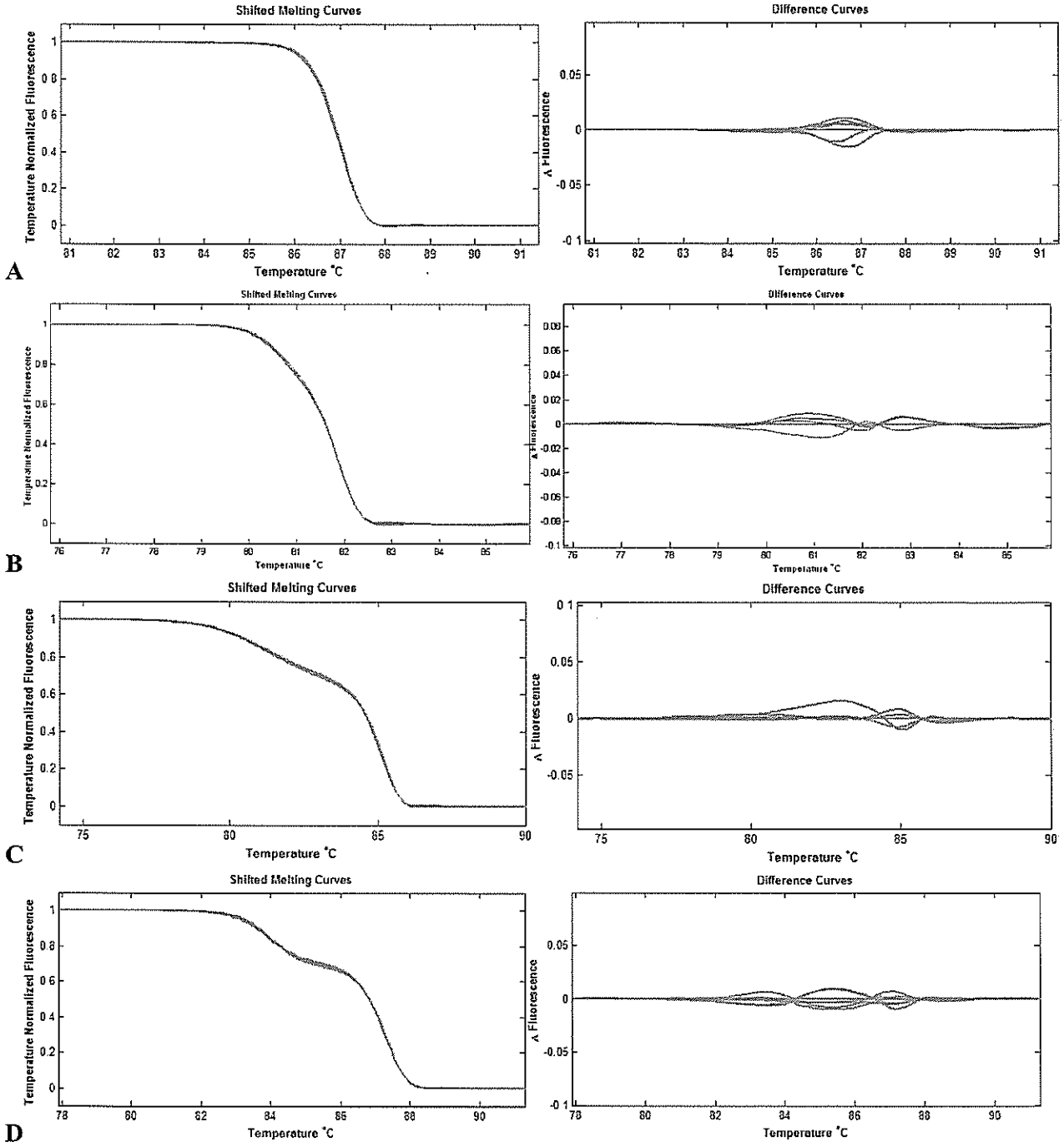
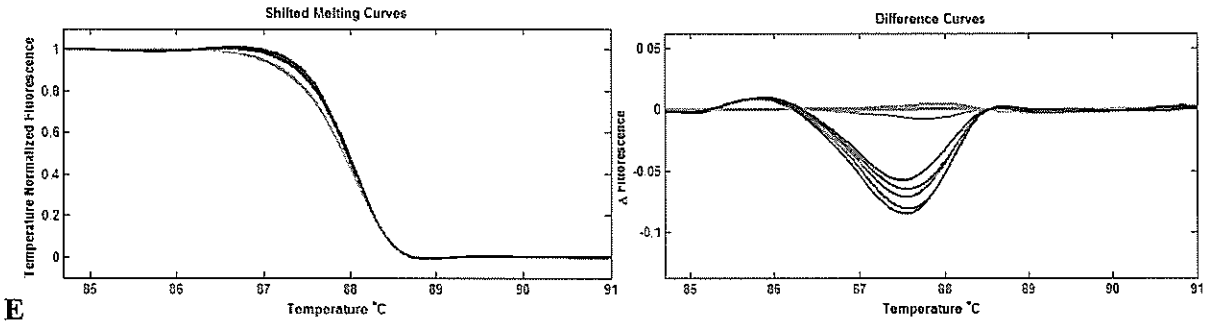
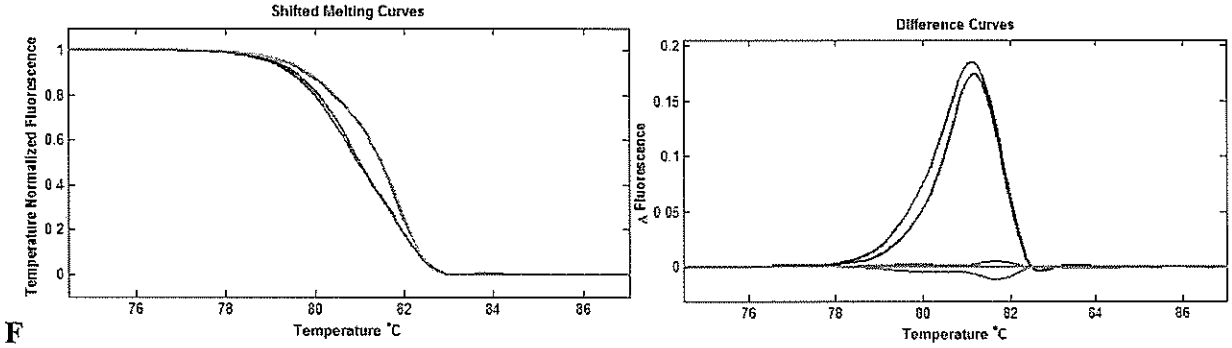

Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
A
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
B
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
C
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
D

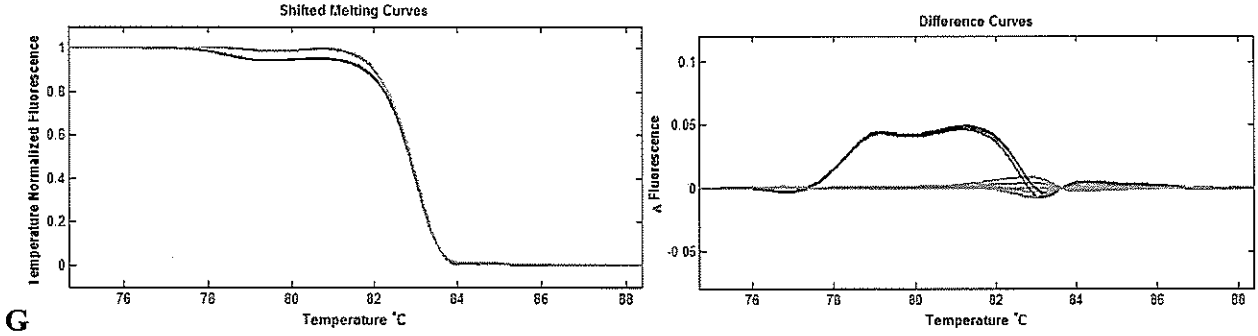
FIGURE 5



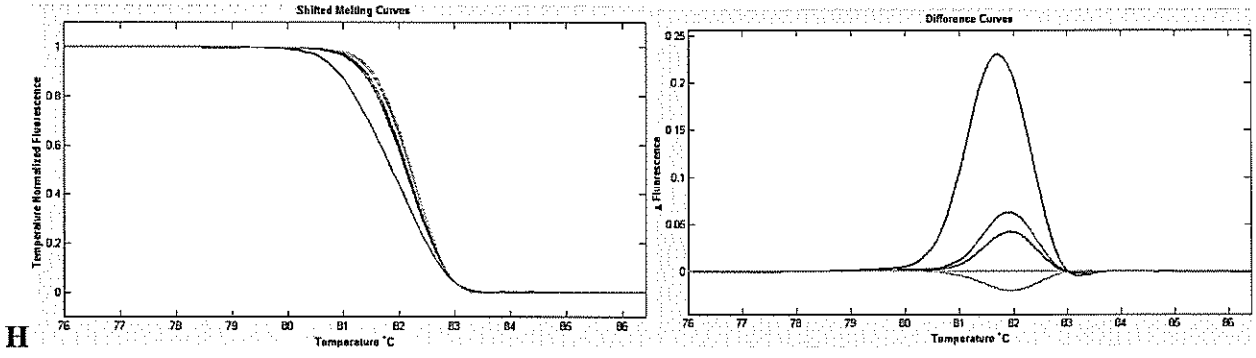
E



F



G



H

FIGURE 6

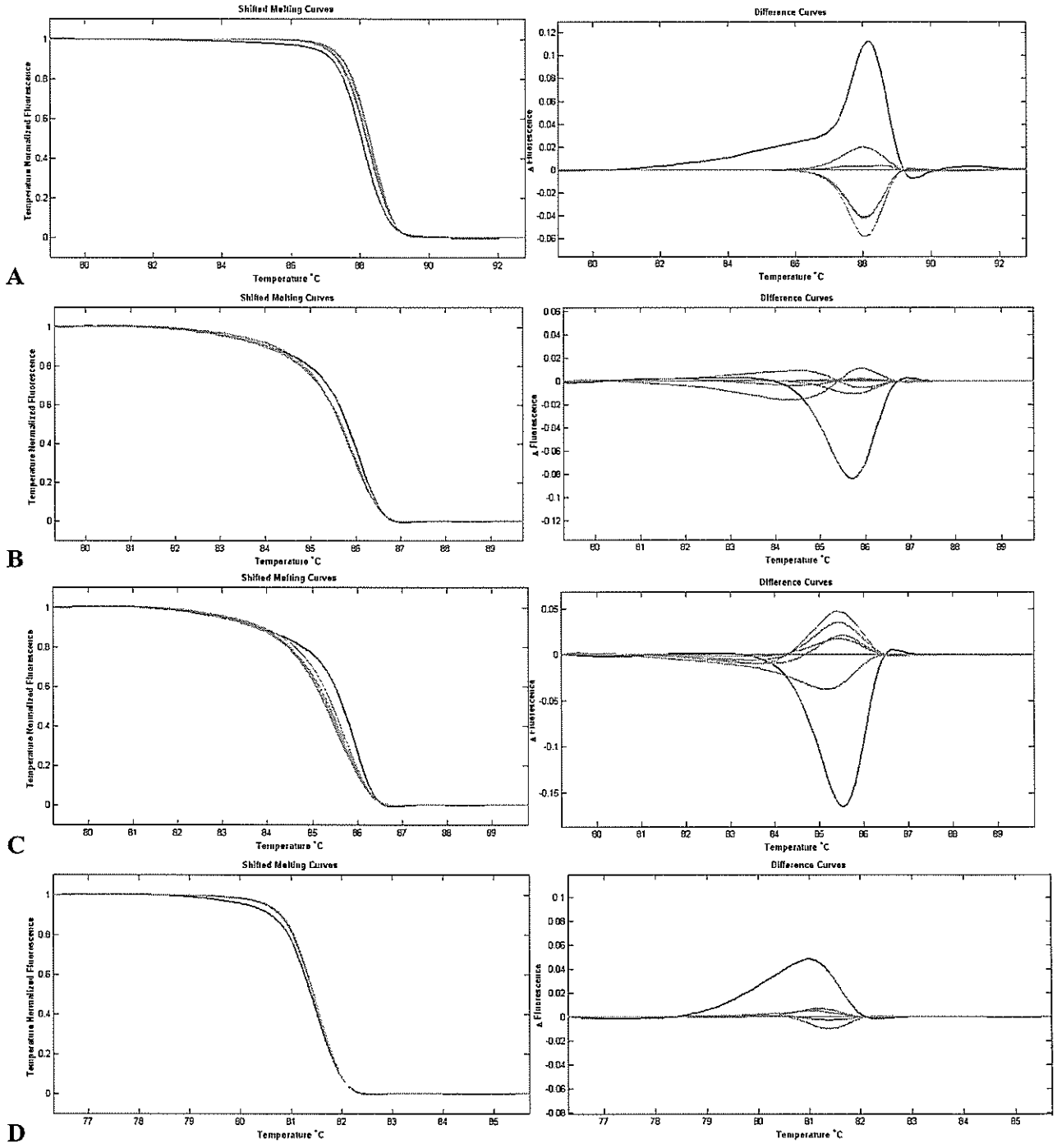

A
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
B
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
C
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
D
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C

FIGURE 6

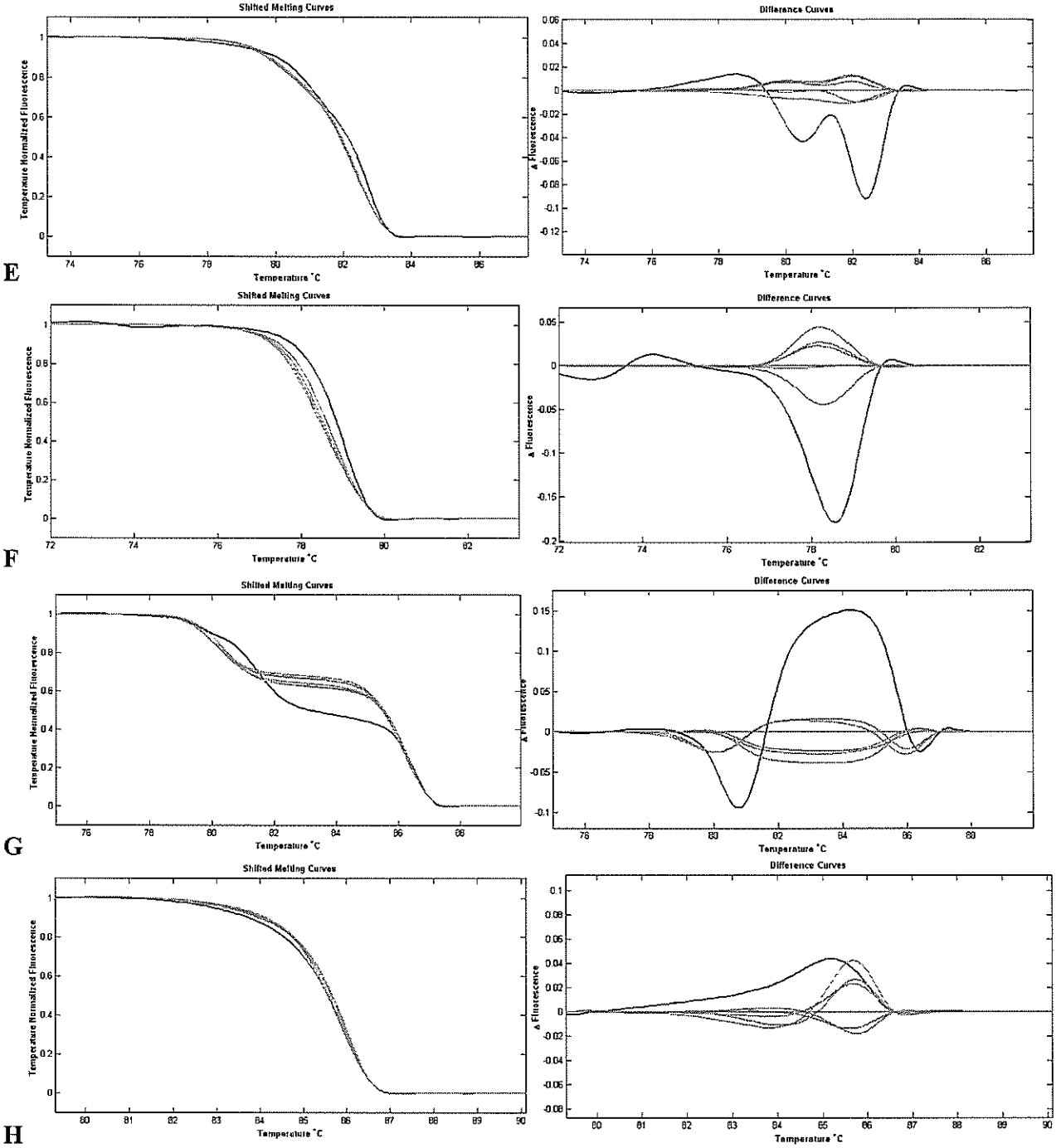

Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
E
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
F
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
G
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
H

FIGURE 6

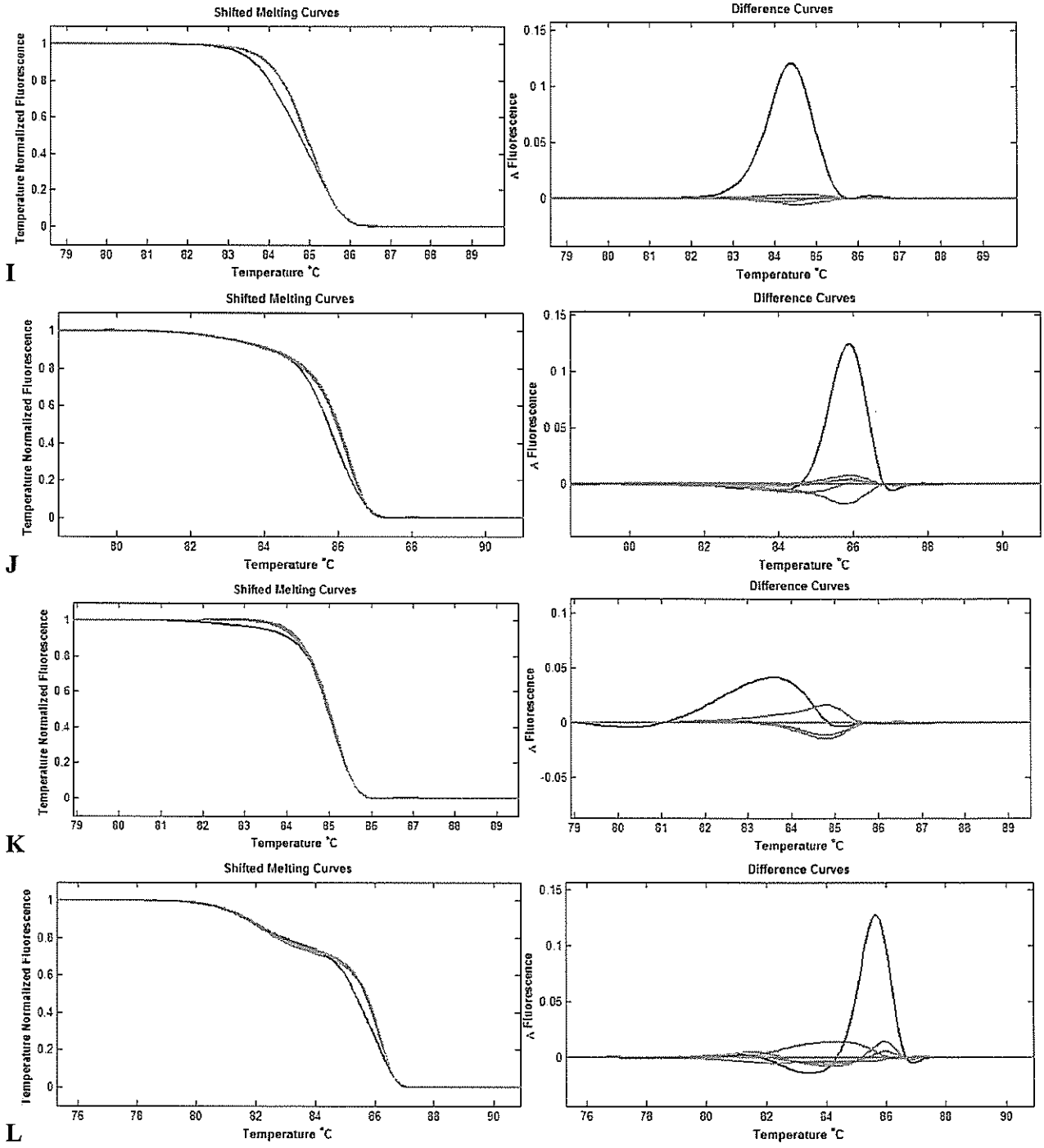

Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
I
J
K
L

FIGURE 6

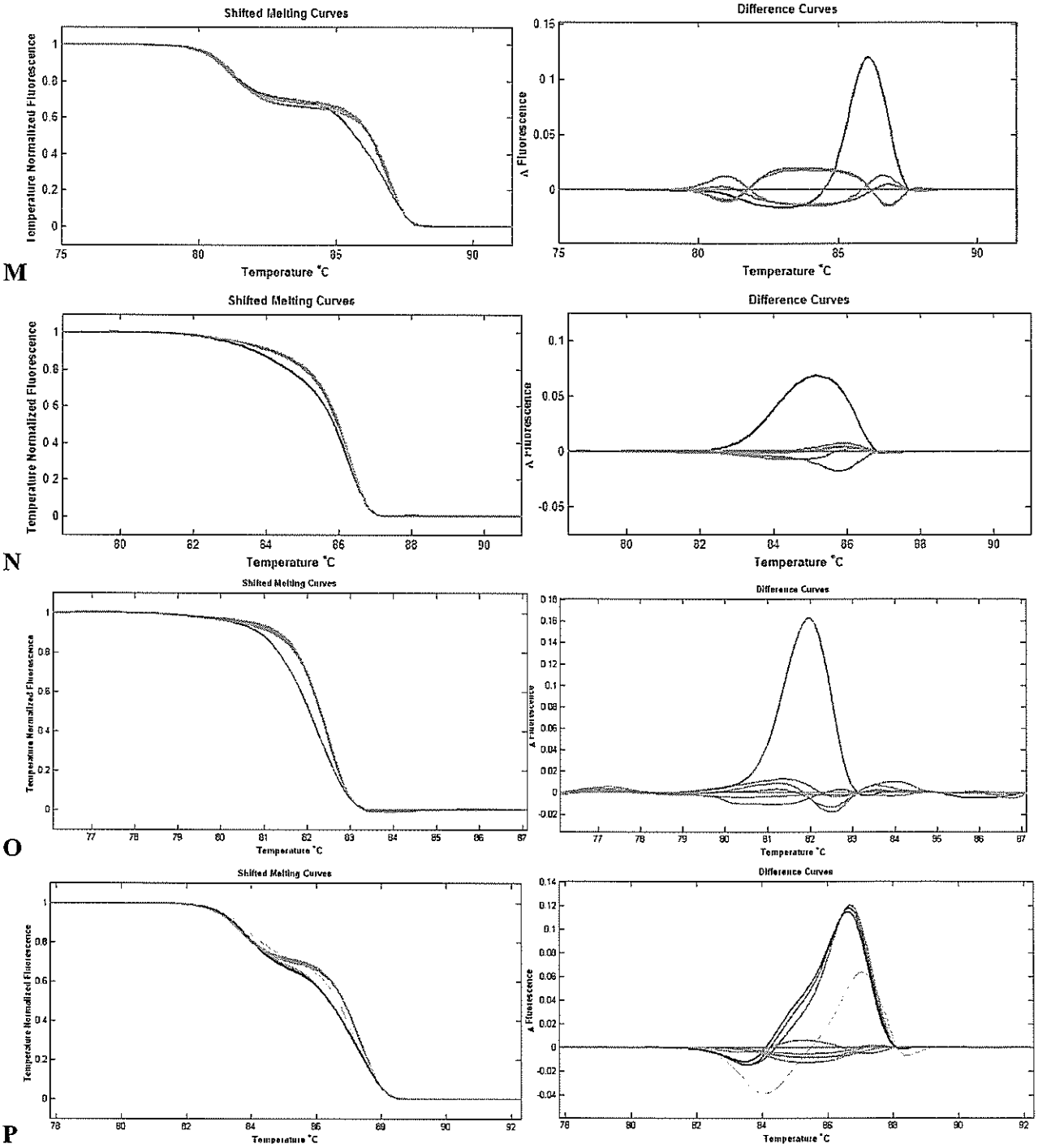
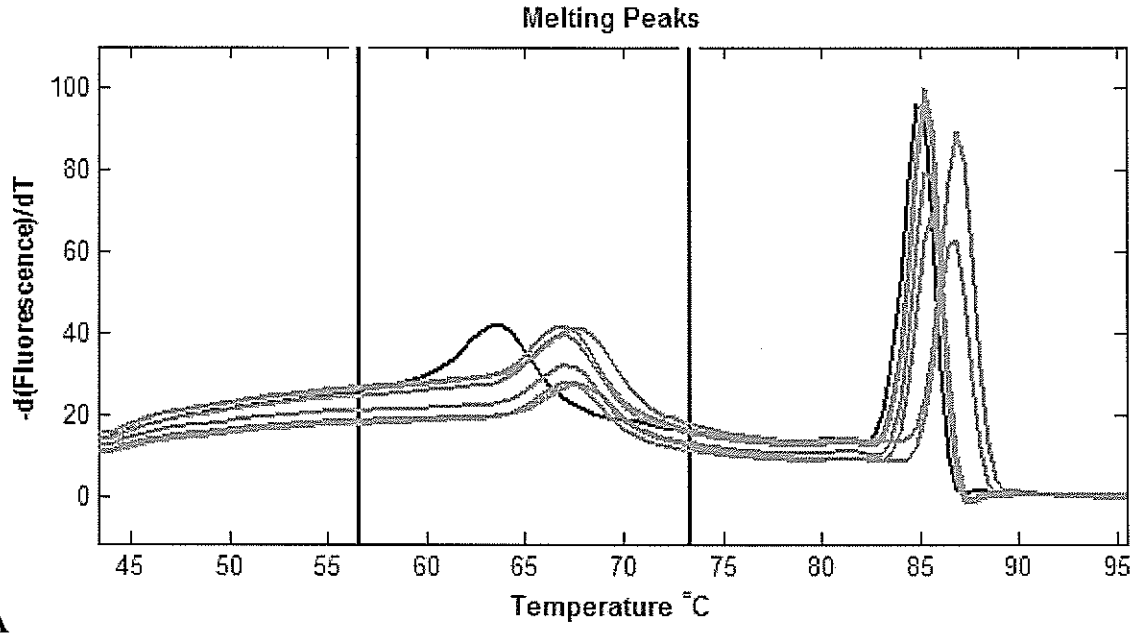
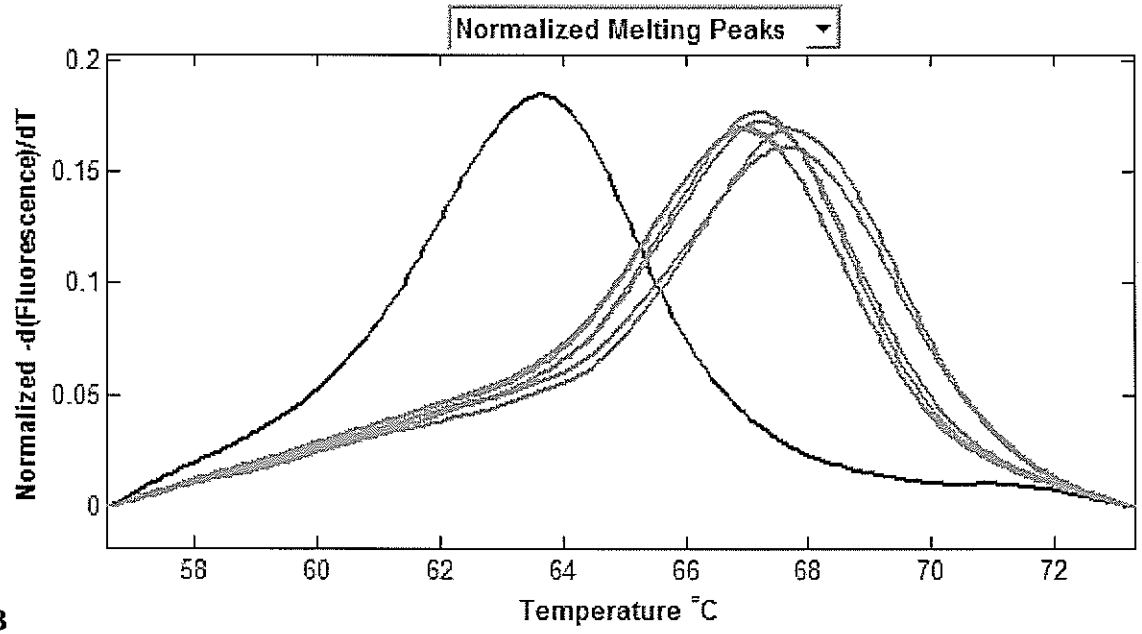

Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
M
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
N
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
O
Shifted Melting Curves
Temperature Normalized Fluorescence
Temperature °C
Difference Curves
Δ Fluorescence
Temperature °C
P

FIGURE 7


Melting Peaks
-d(Fluorescence)/dT
100
80
60
40
20
0
45
50
55
60
65
70
75
80
85
90
95
Temperature °C

A


Normalized Melting Peaks
Normalized -d(Fluorescence)/dT
0.2
0.15
0.1
0.05
0
58
60
62
64
66
68
70
72
Temperature °C

B