RAMA UNIVERSITY

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FACULTY OF ENGINEERING &TECHNOLOGY
DEPARTMENT OF BIOTECHNOLOGY

- 3.This mixture is kept in a PCR equipment and is subjected to repeated cycles of DNA denaturation-renaturation-DNA replication.

- 4.During this process, the decaoligonucleotide will pair with the homologous sequence present at different locations in the DNA.

- 5.DNA replication extend the decaoligonucleotide and copy the sequence continuous with the sequence with which the selected oligonucleotide has paired.

- 6.The repeated cycles of denaturation-renaturation-DNA replication will amplify this sequence of DNA.

- 7.Amplification will takes place only of those regions of the genome that has the sequence complementary to the decaoligonucleotide at their both ends.

- 8. After several cycles of amplification the DNA is subjected to gel electrophoresis.

Initial denaturation at 94°C for 10 min.

↓

Denaturation at 94°C for 1 min. Annealing at 37-45°C for 1 min. Extension at 72°C for 1 min.

↓

Final extension at 72°C for 10 min.

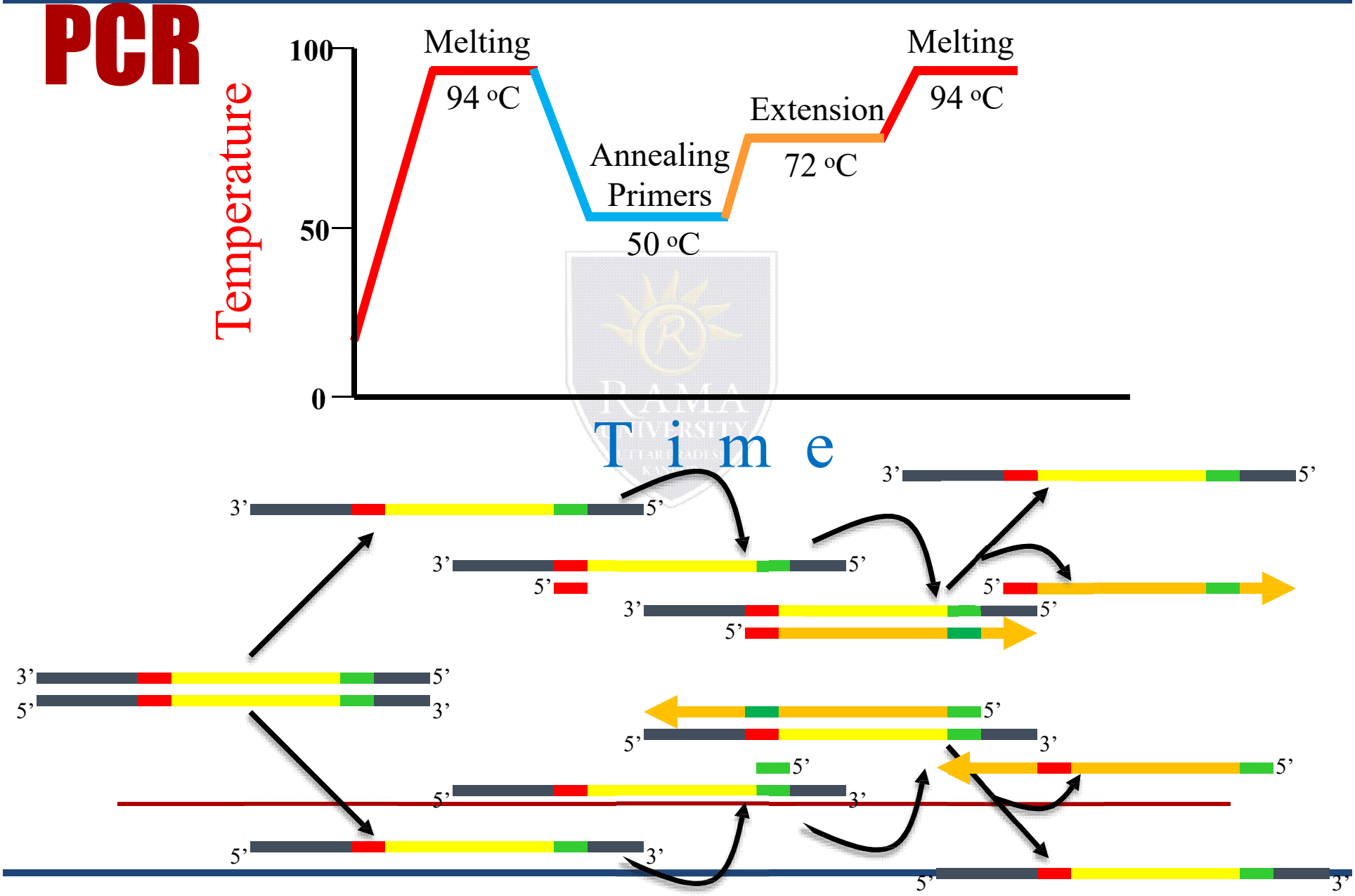
↓

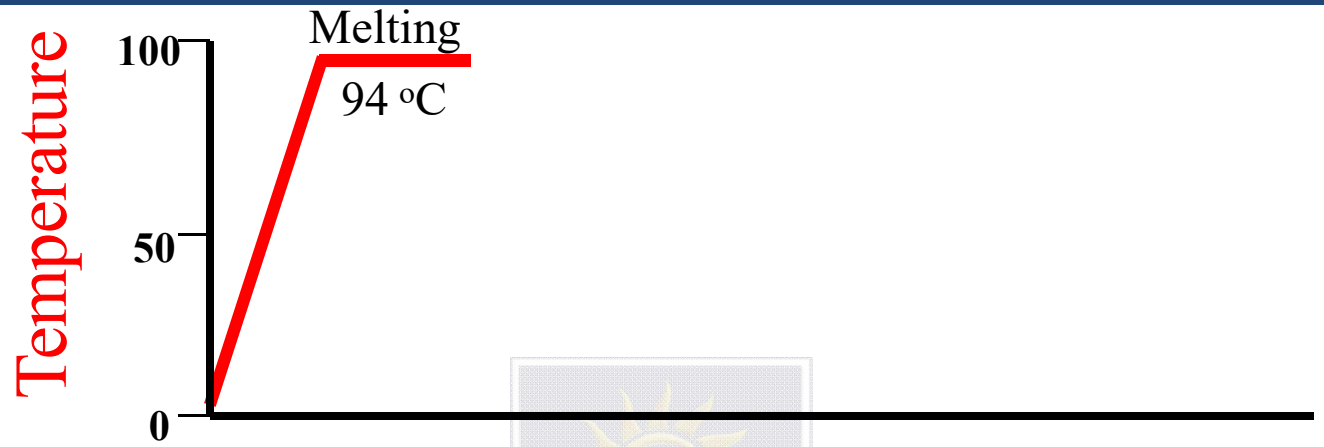
Cooling at 4°C.

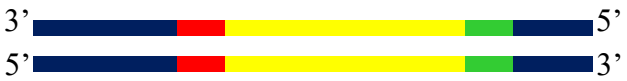
↓

- 9.The amplified DNA will form a distinct band. it is detected by ethidium bromide staining and visible fluorescence's under U.V. light

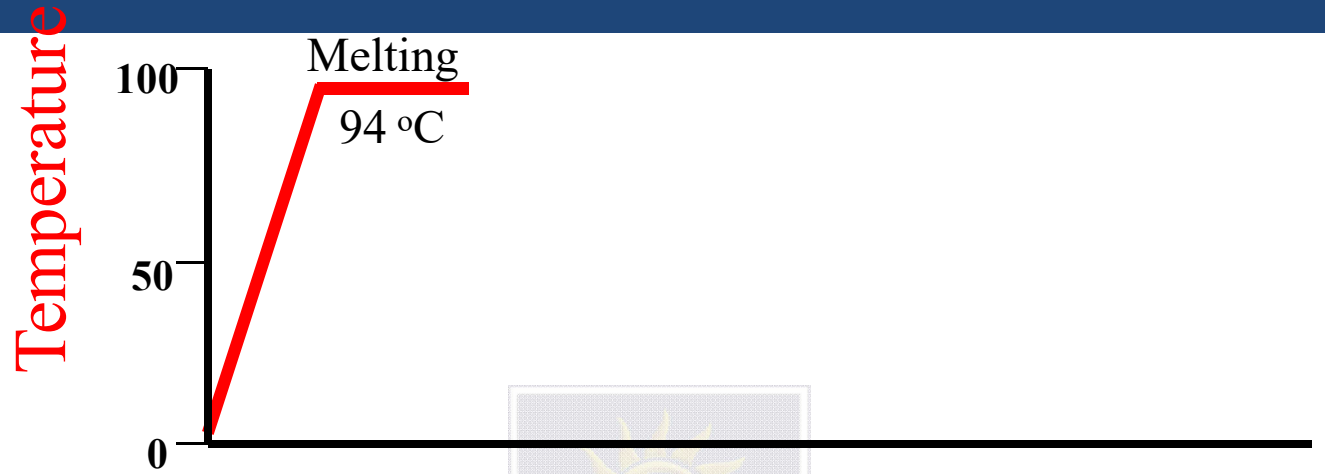
PCR

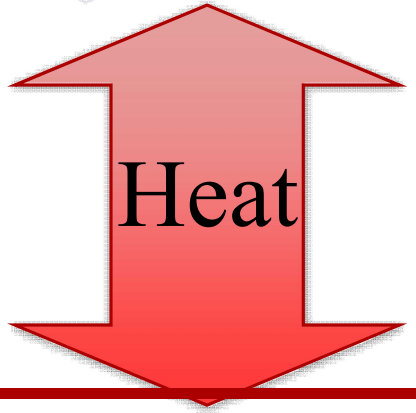

Temperature
100
50
0
Melting
94 °C
Annealing
Primers
50 °C
Extension
72 °C
Melting
94 °C
Time
3'
5'



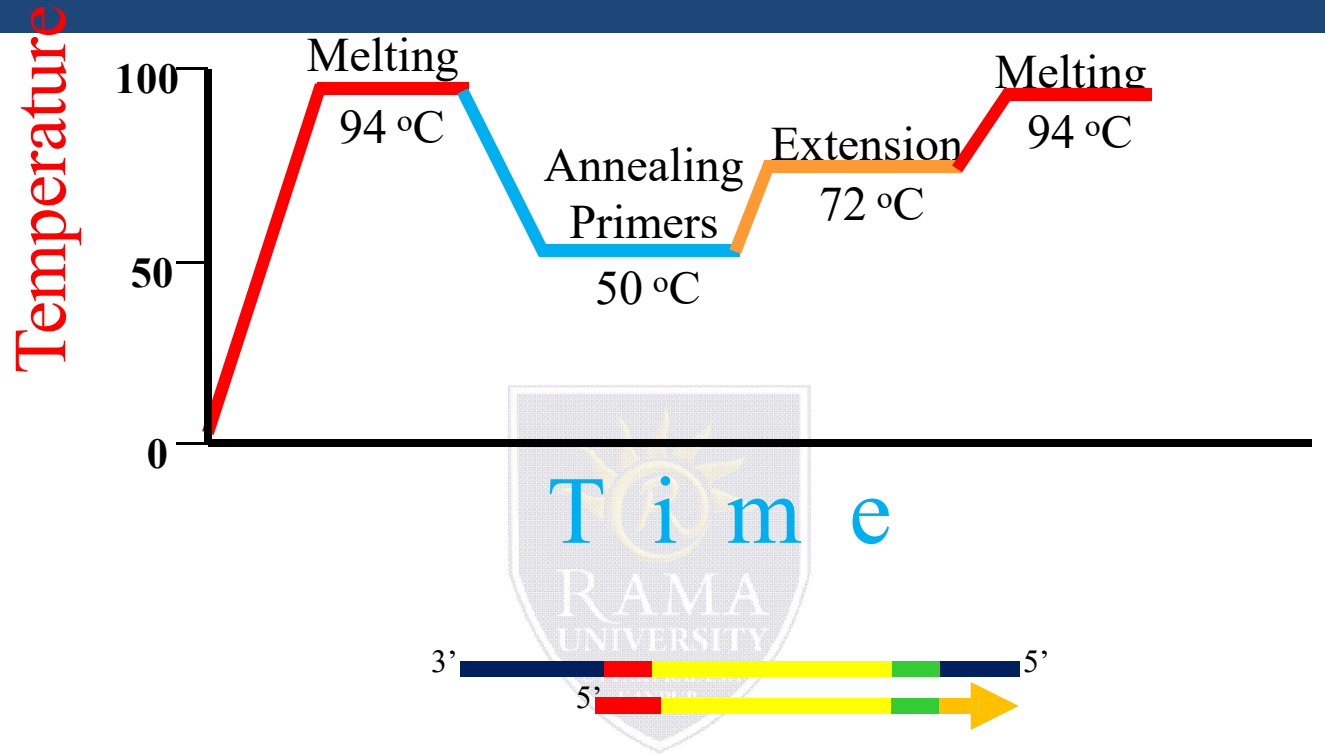


RAPD



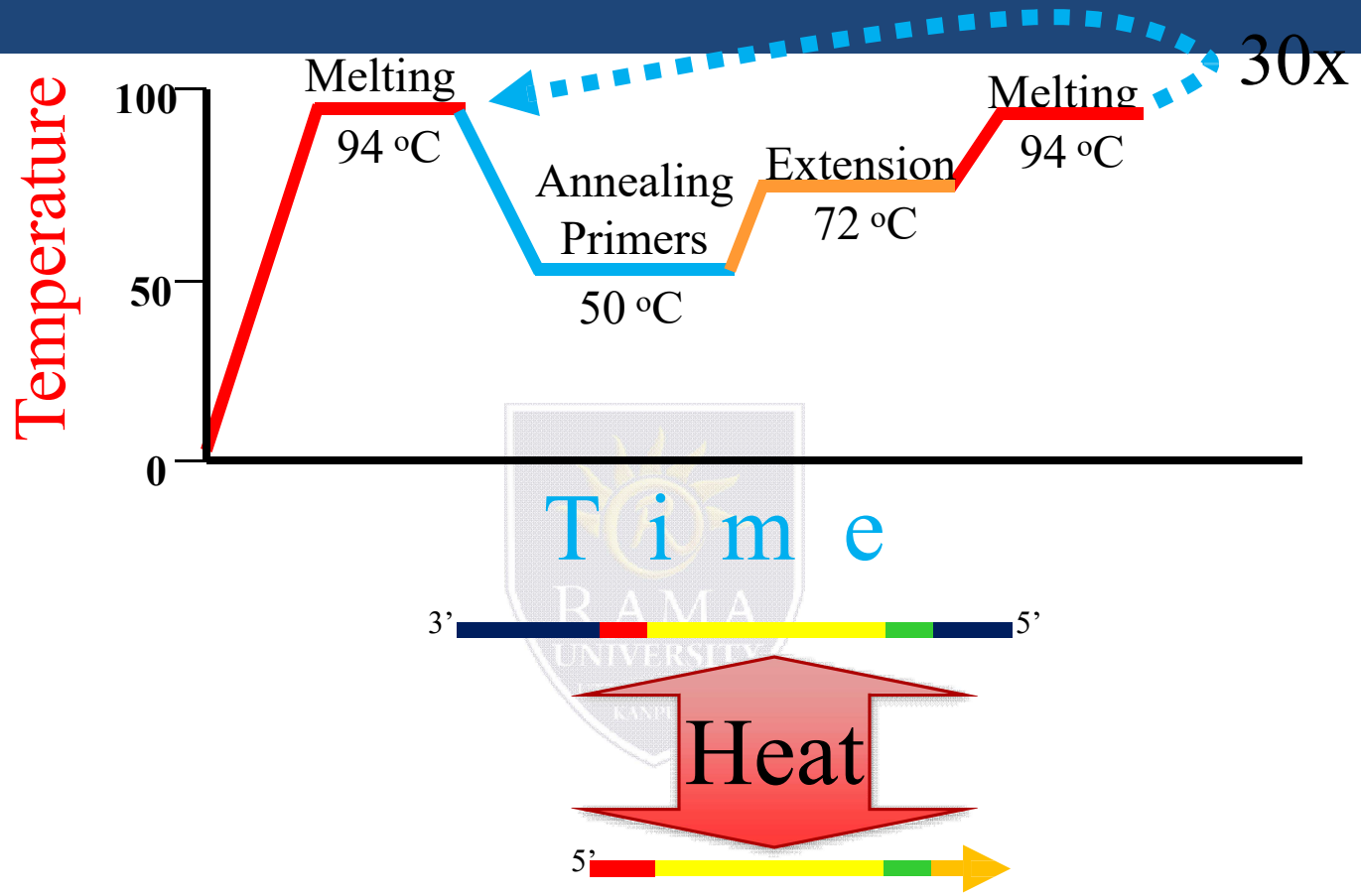


RAPD


Temperature
100
50
0
Melting
94 °C
Annealing
Primers
50 °C
Extension
72 °C
Melting
94 °C
Time
3'
5'
5'

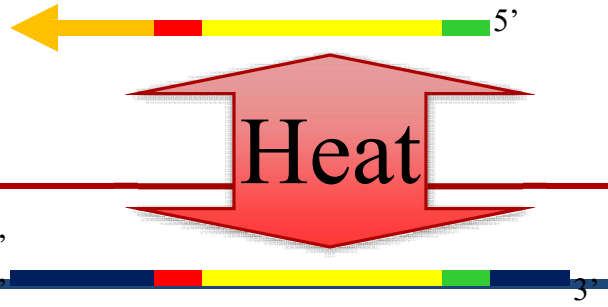
RAPD

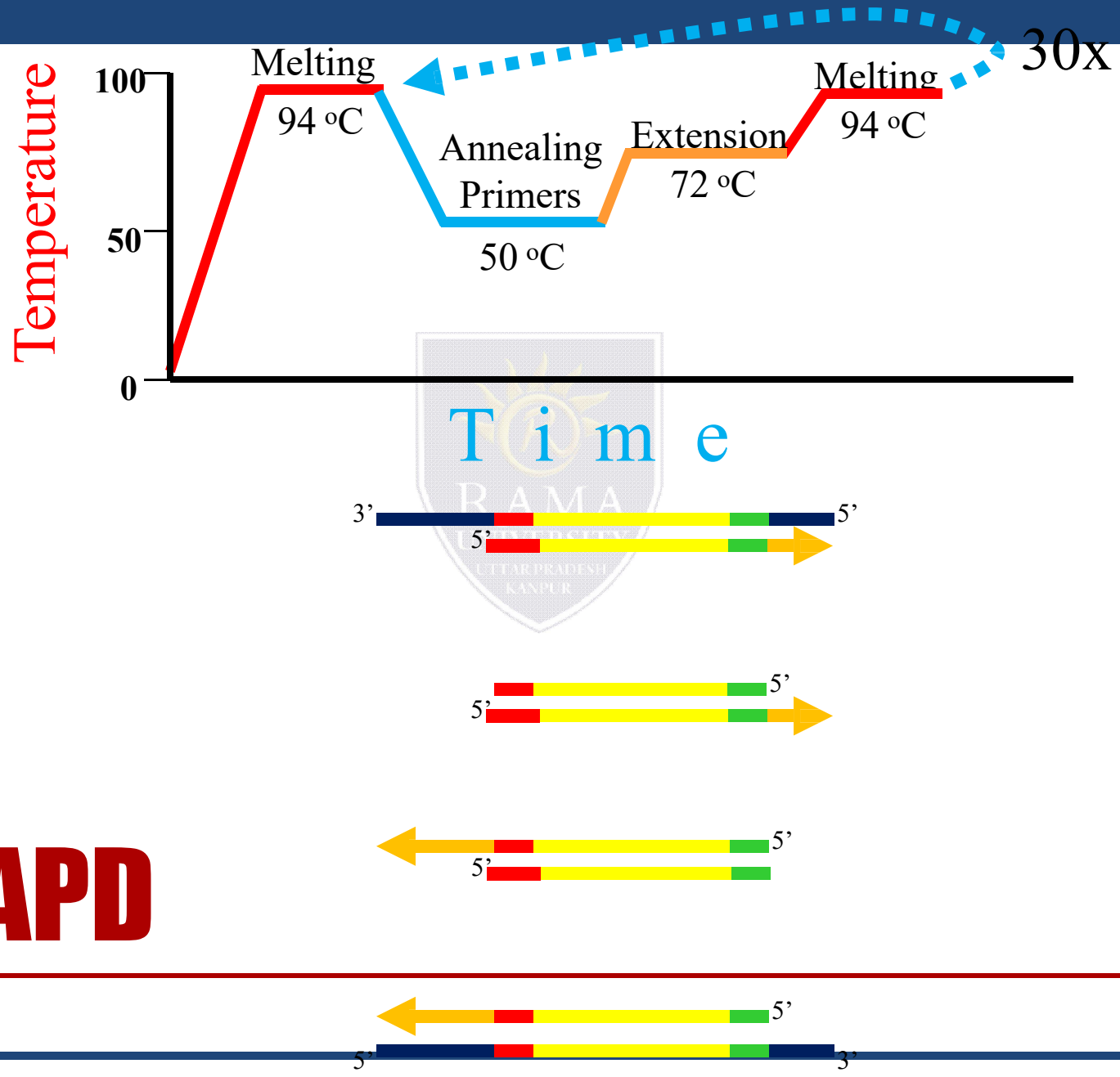

5'
5'
3'


Temperature
100
50
0
Melting
94 °C
Annealing
Primers
50 °C
Extension
72 °C
Melting
94 °C
30x
Time

3'
5'
Heat
5'

RAPD


5'
Heat
5'
5'
3'


Temperature
100
50
0
Melting
94 °C
Annealing
Primers
50 °C
Extension
72 °C
Melting
94 °C
30x
Time
3'
5'
5'
5'
5'
5'
5'
5'
5'
3'

RAPD