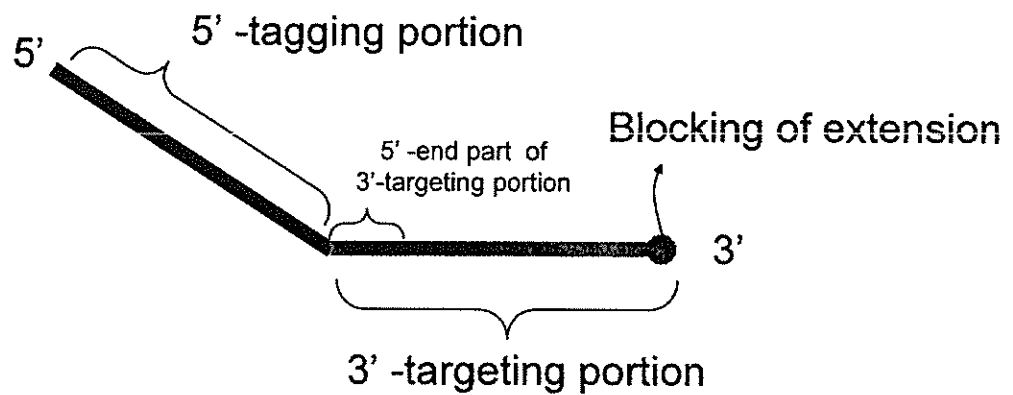
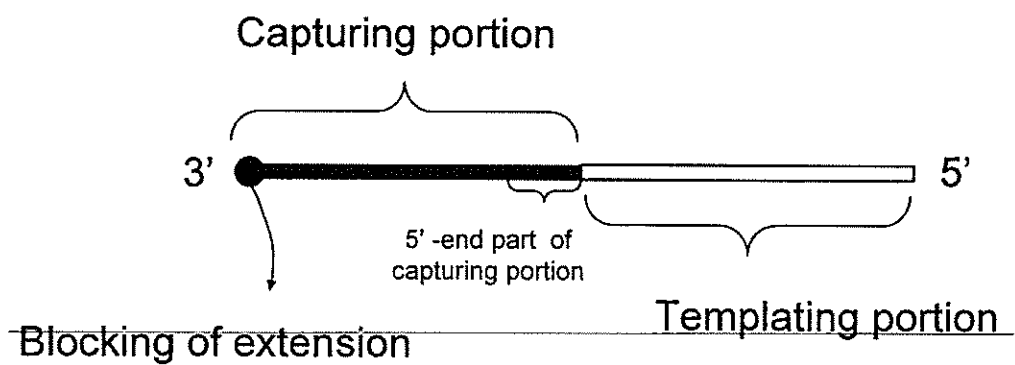


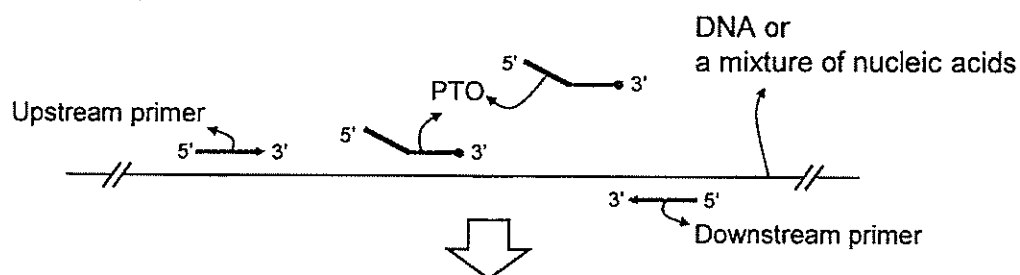
1/30

Fig. 1**A. Probing and Tagging Oligonucleotide (PTO)****B. Capturing and Templating Oligonucleotide (CTO)**

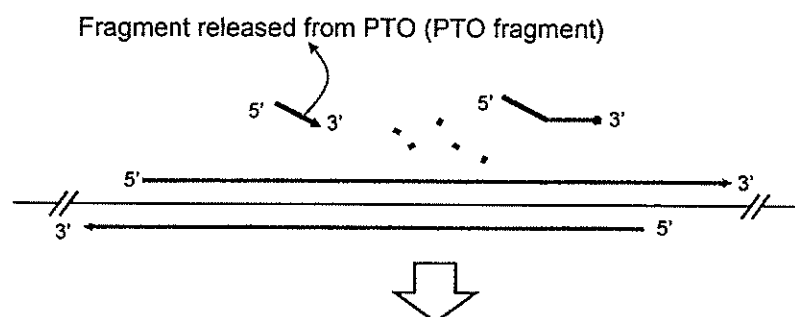
2/30

Fig. 2

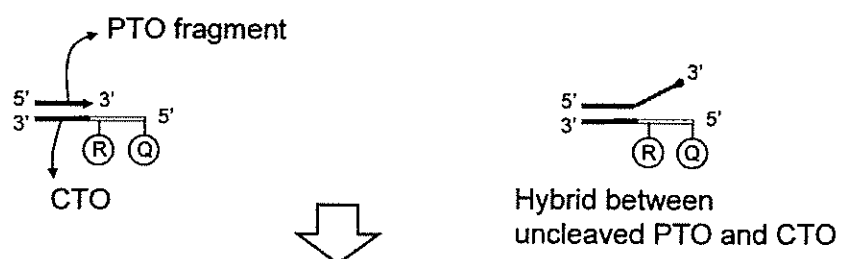
A. Hybridization



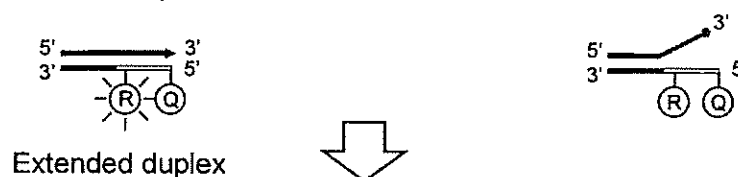
B. Primer extension & Cleavage of PTO



C. Hybridization of PTO fragment to CTO



D. Extension of PTO fragment



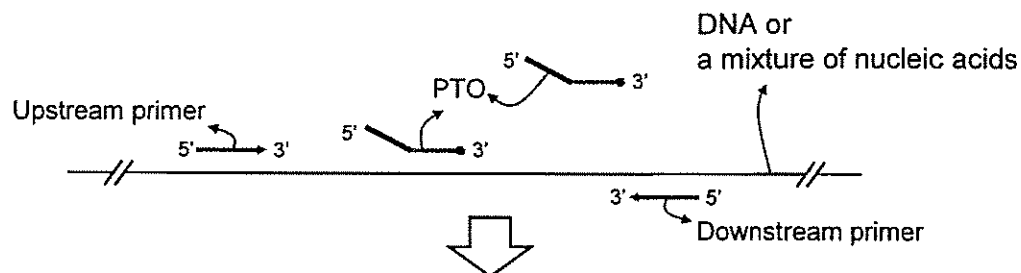
E. Melting analysis



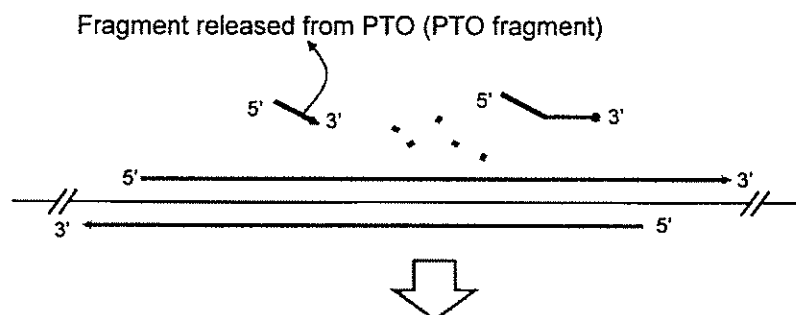
3/30

Fig. 3

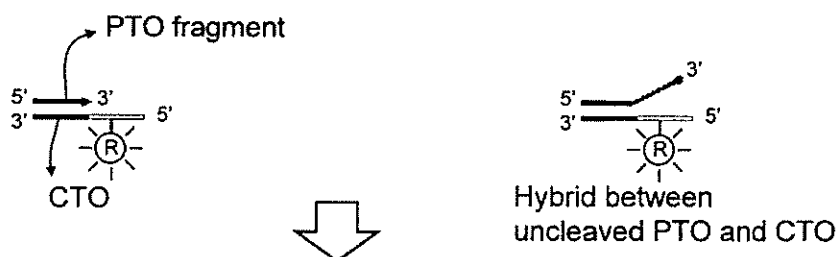
A. Hybridization



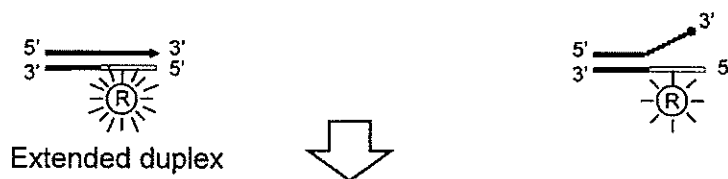
B. Primer extension & Cleavage of PTO



C. Hybridization of PTO fragment to CTO



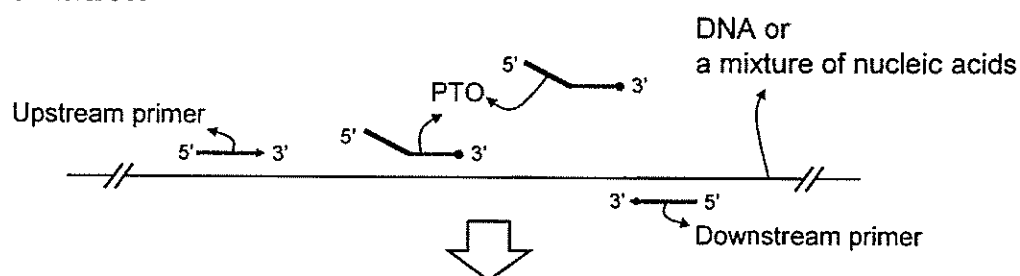
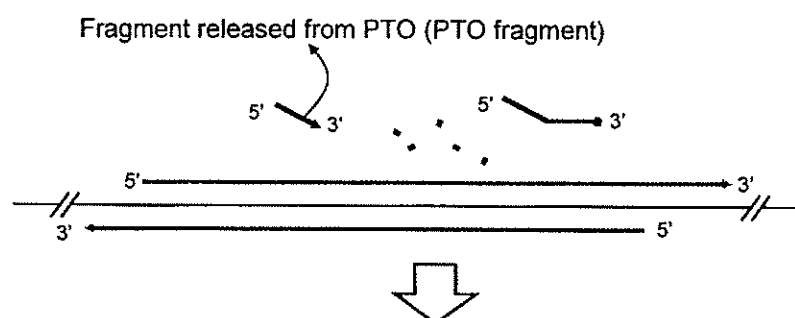
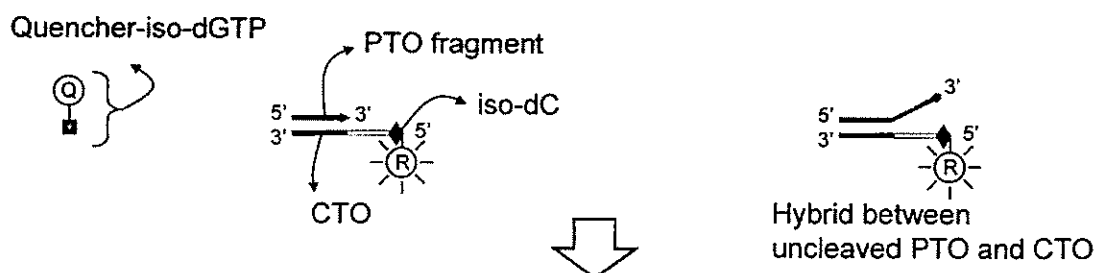
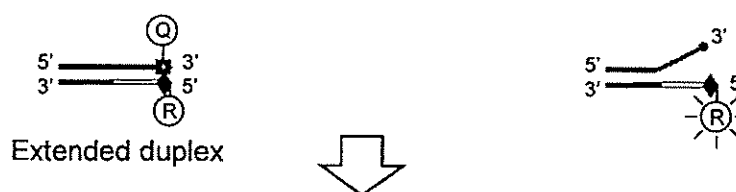
D. Extension of PTO fragment



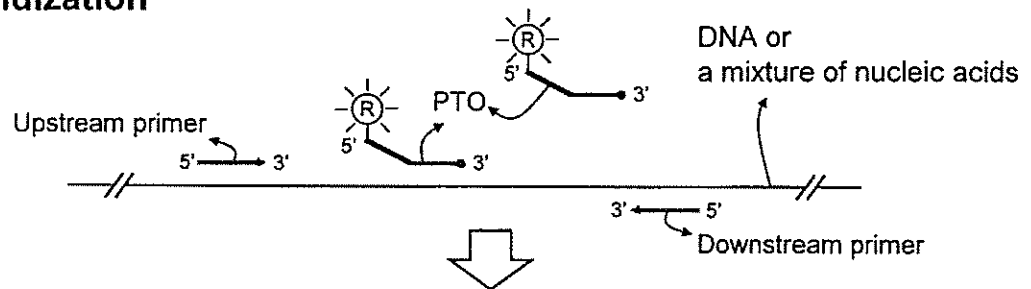
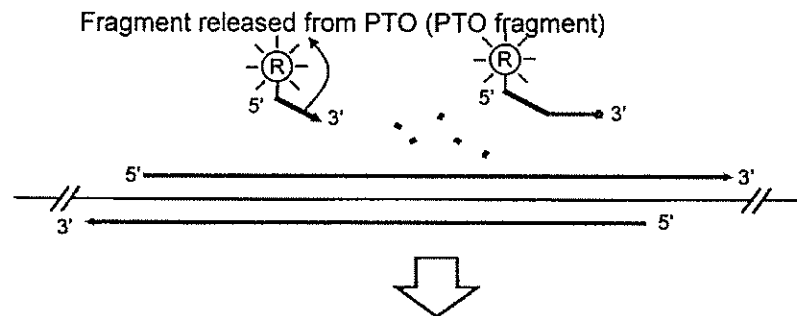
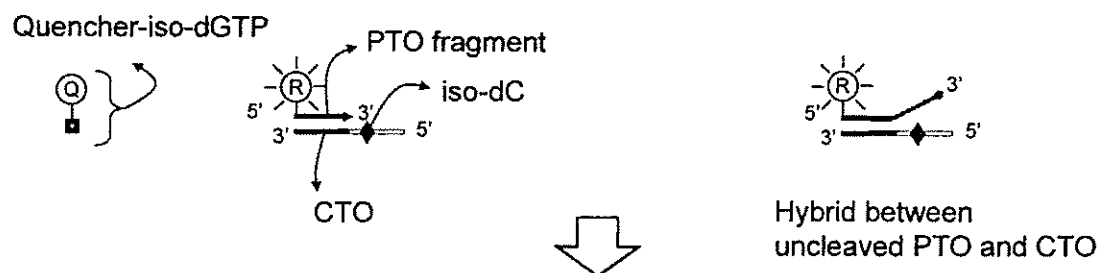
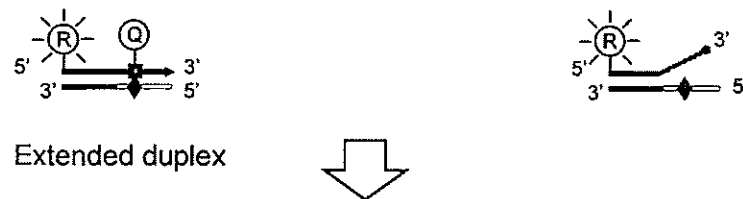
E. Melting analysis



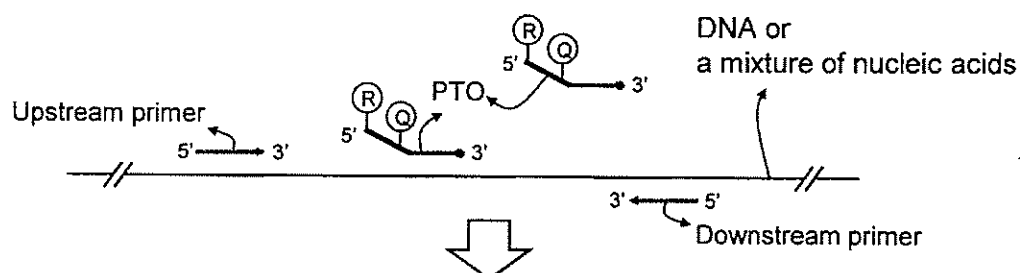
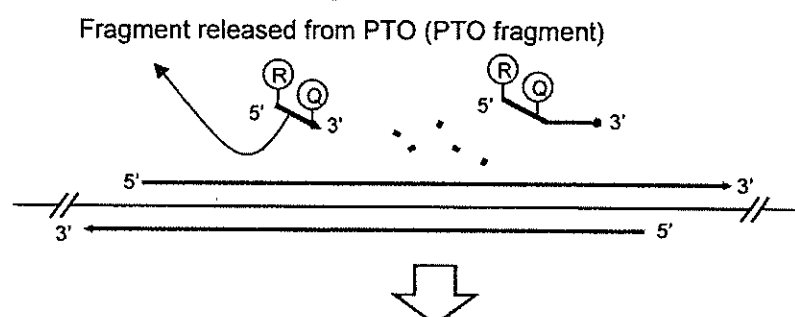
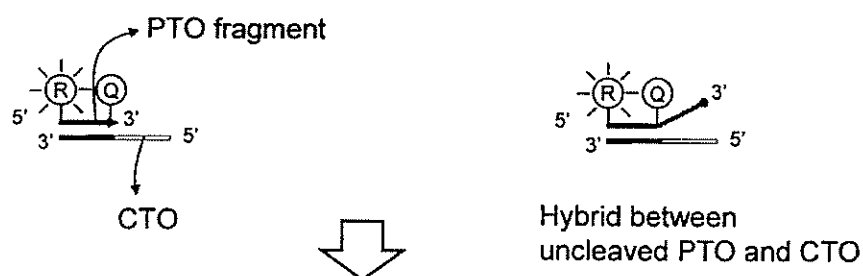
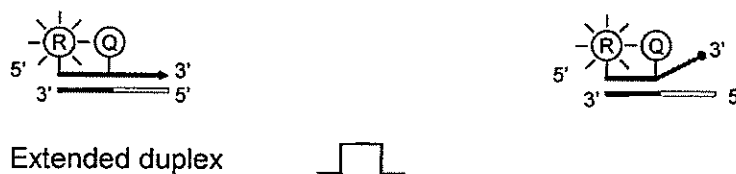
4/30

Fig. 4**A. Hybridization****B. Primer extension & Cleavage of PTO****C. Hybridization of PTO fragment to CTO****D. Extension of PTO fragment****E. Melting analysis**

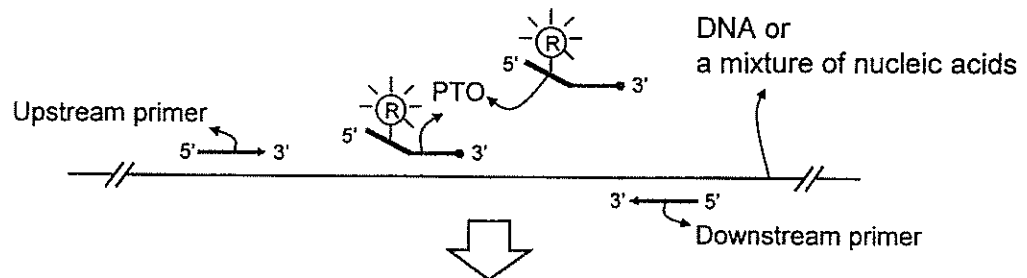
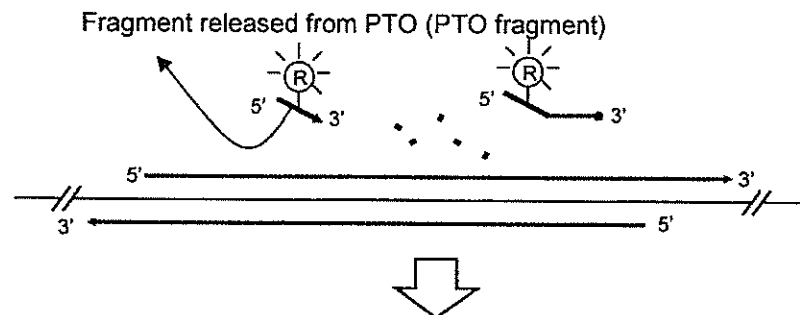
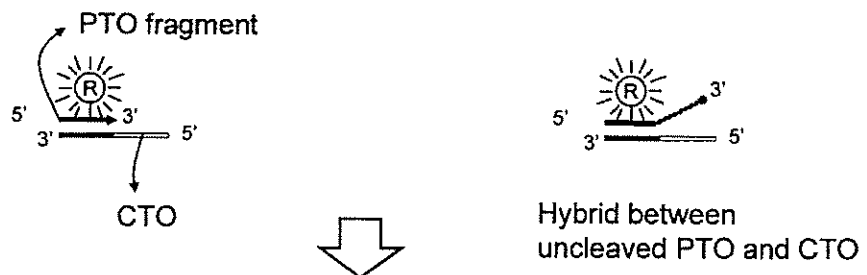
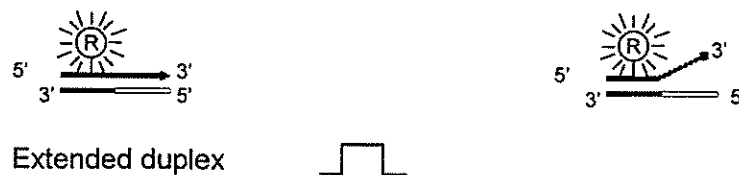
5/30

Fig. 5**A. Hybridization****B. Primer extension & Cleavage of PTO****C. Hybridization of PTO fragment to CTO****D. Extension of PTO fragment****E. Melting analysis**

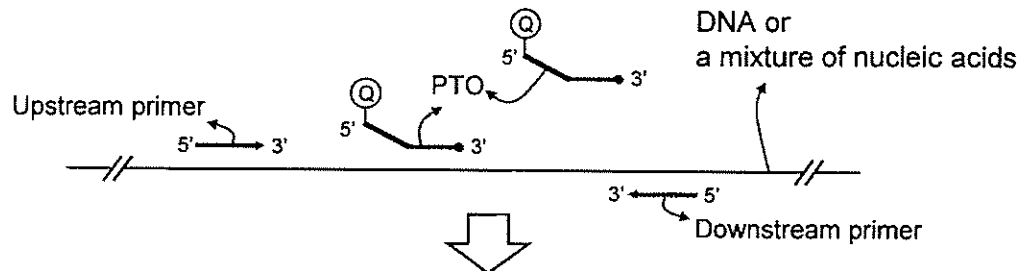
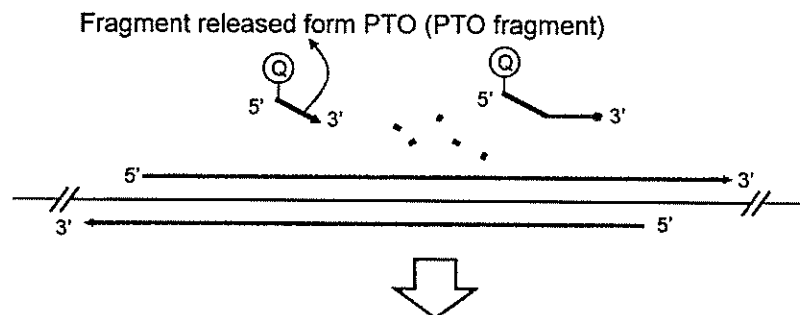
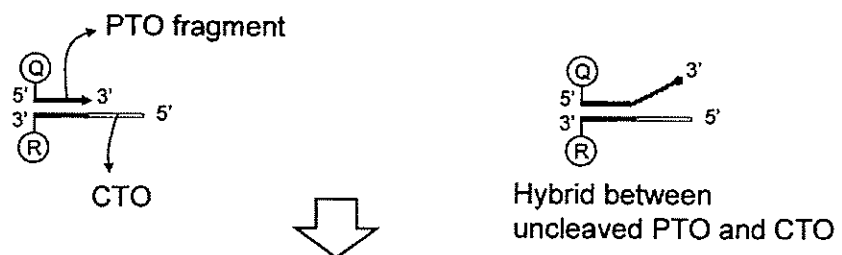
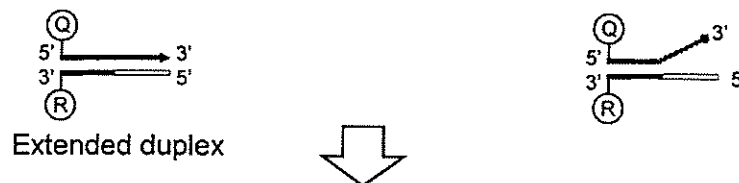
6/30

Fig. 6**A. Hybridization****B. Primer extension & Cleavage of PTO****C. Hybridization of PTO fragment to CTO****D. Extension of PTO fragment****E. Melting analysis**

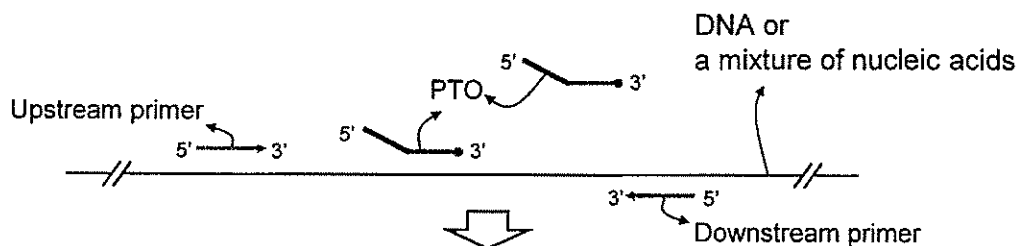
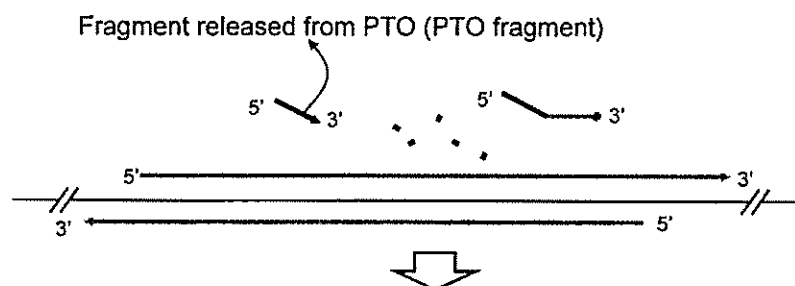
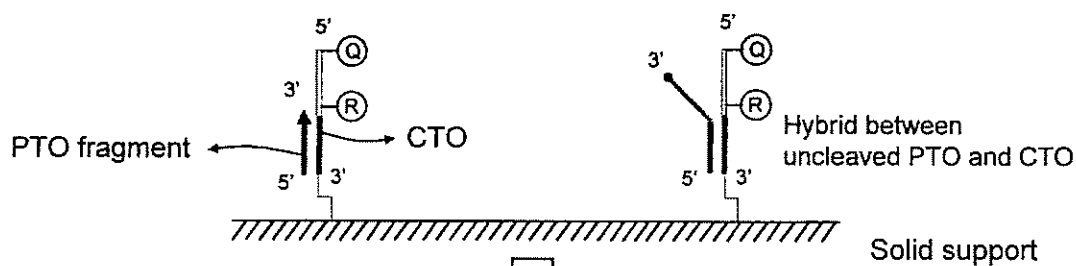
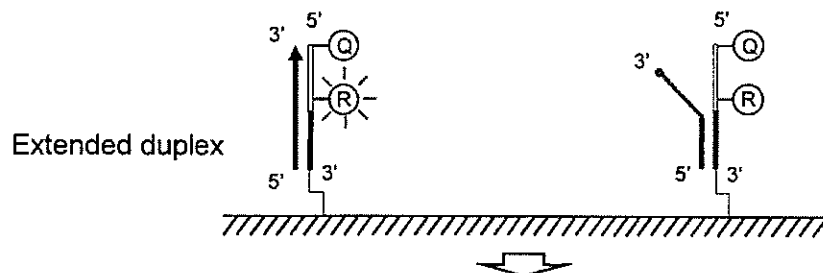
7/30

Fig. 7**A. Hybridization****B. Primer extension & Cleavage of PTO****C. Hybridization of PTO fragment to CTO****D. Extension of PTO fragment****E. Melting analysis**

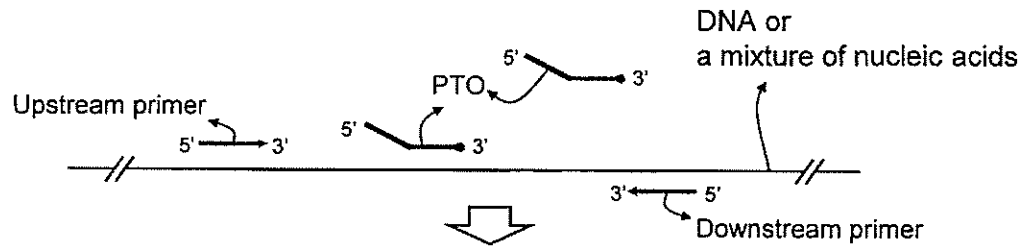
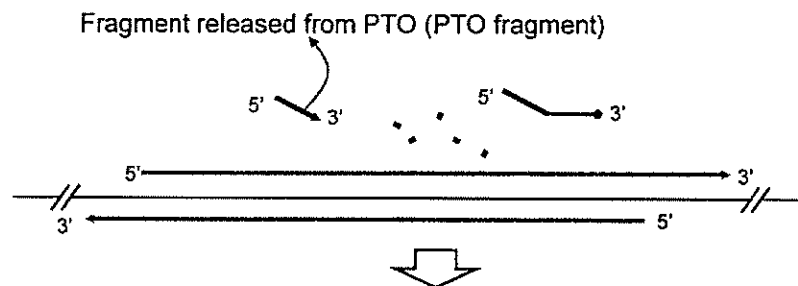
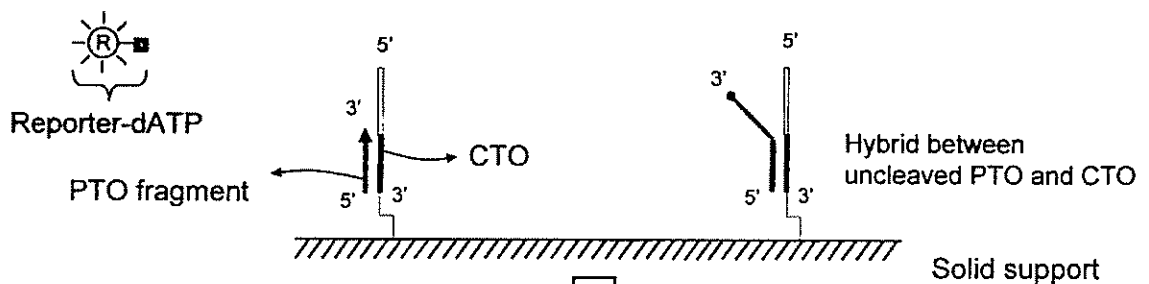
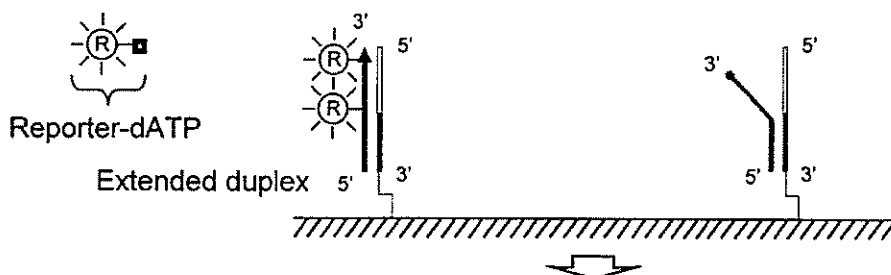
8/30

Fig. 8**A. Hybridization****B. Primer extension & Cleavage of PTO****C. Hybridization of PTO fragment to CTO****D. Extension of PTO fragment****E. Melting analysis**

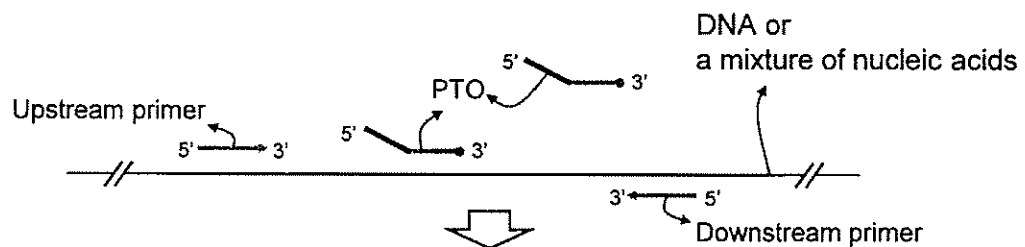
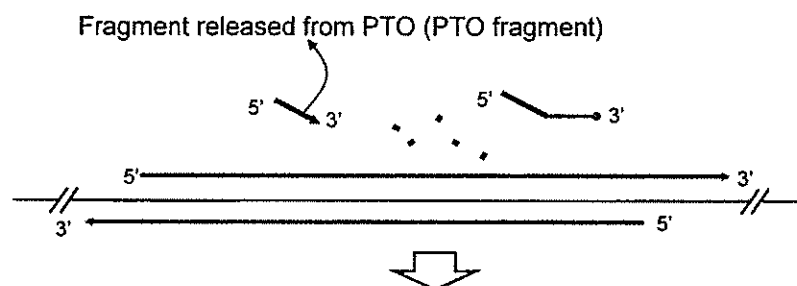
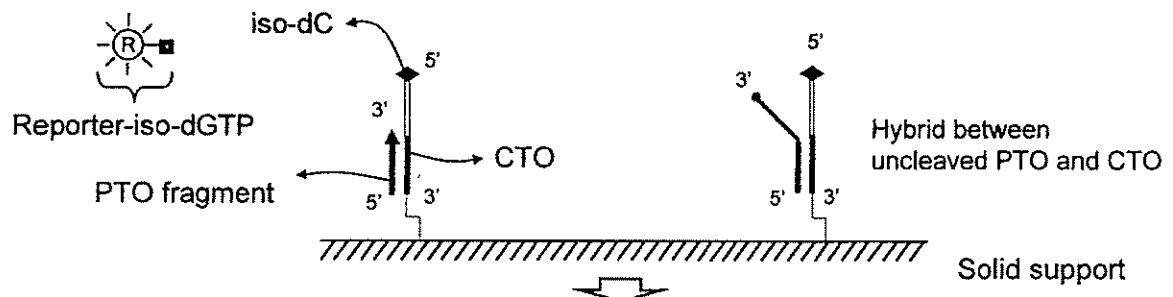
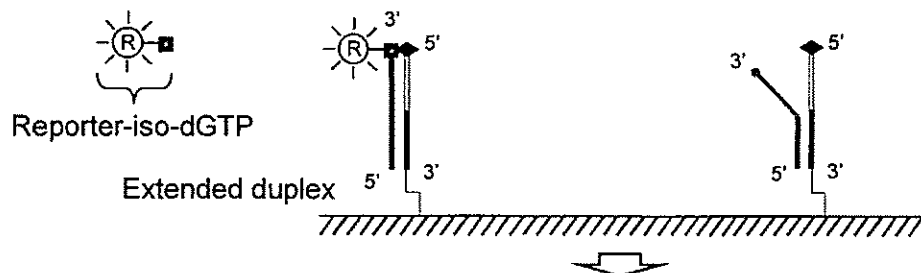
9/30

Fig. 9**A. Hybridization****B. Primer extension & Cleavage of PTO****C. Hybridization of PTO fragment to CTO****D. Extension of PTO fragment****E. Detection at a pre-determined temperature**

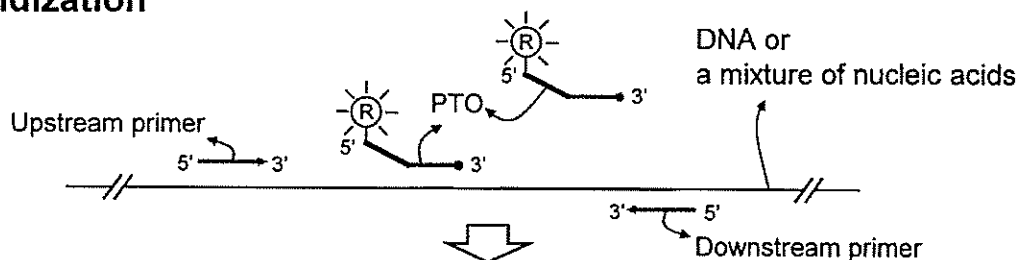
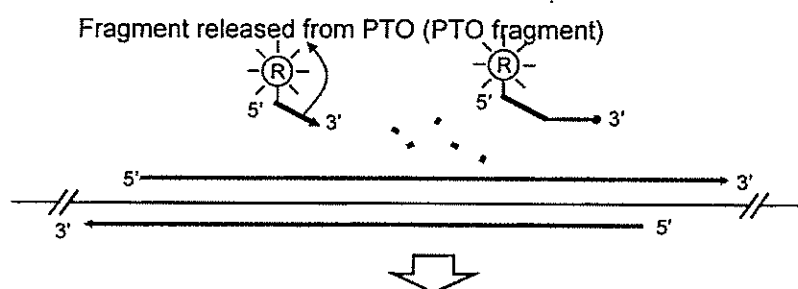
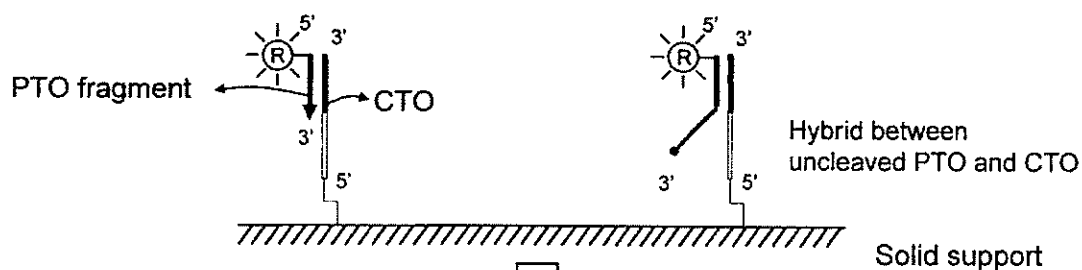
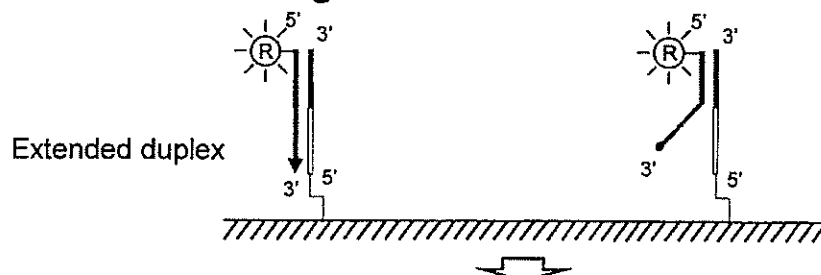
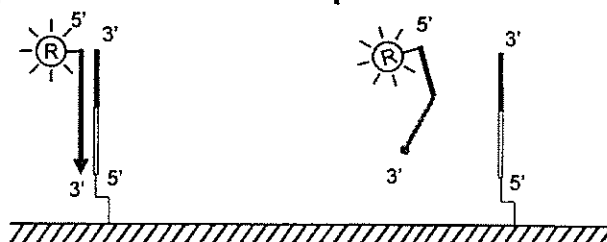
10/30

Fig. 10**A. Hybridization****B. Primer extension & Cleavage of PTO****C. Hybridization of PTO fragment to CTO****D. Extension of PTO fragment****E. Detection at a pre-determined temperature**

11/30

Fig. 11**A. Hybridization****B. Primer extension & Cleavage of PTO****C. Hybridization of PTO fragment to CTO****D. Extension of PTO fragment****E. Detection at a pre-determined temperature**

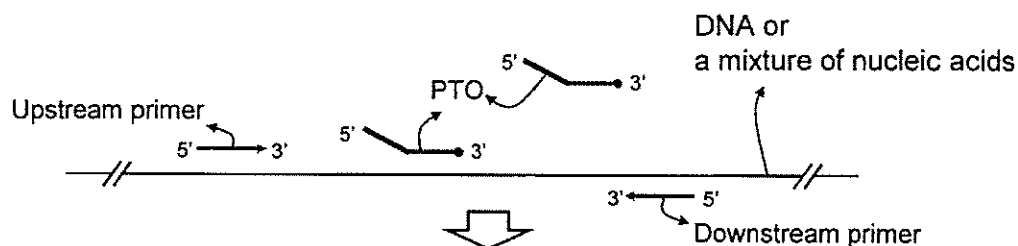
12/30

Fig. 12**A. Hybridization****B. Primer extension & Cleavage of PTO****C. Hybridization of PTO fragment to CTO****D. Extension of PTO fragment****E. Detection at a pre-determined temperature**

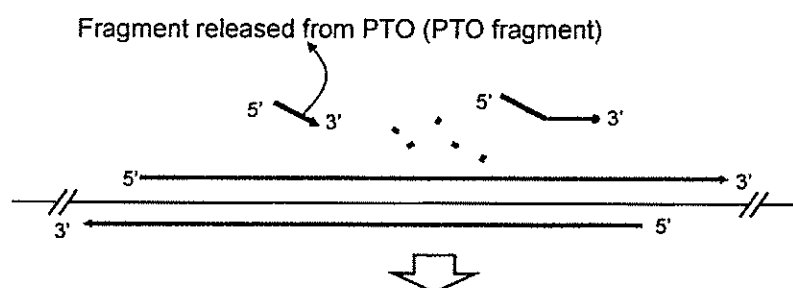
13/30

Fig. 13

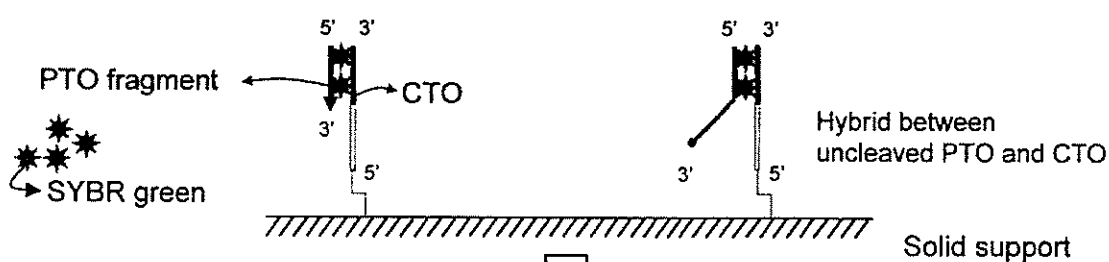
A. Hybridization



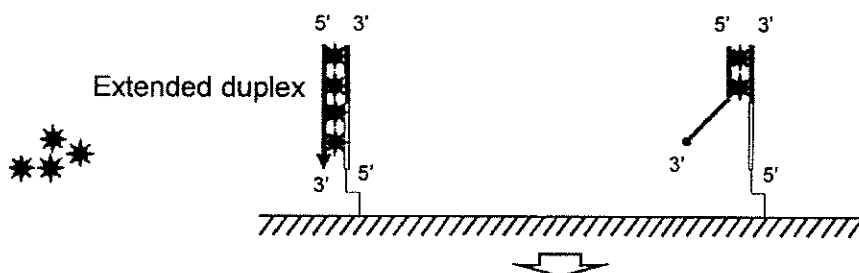
B. Primer extension & Cleavage of PTO



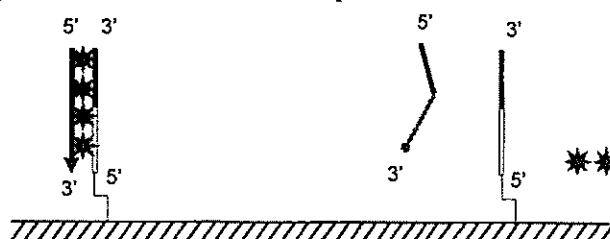
C. Hybridization of PTO fragment to CTO



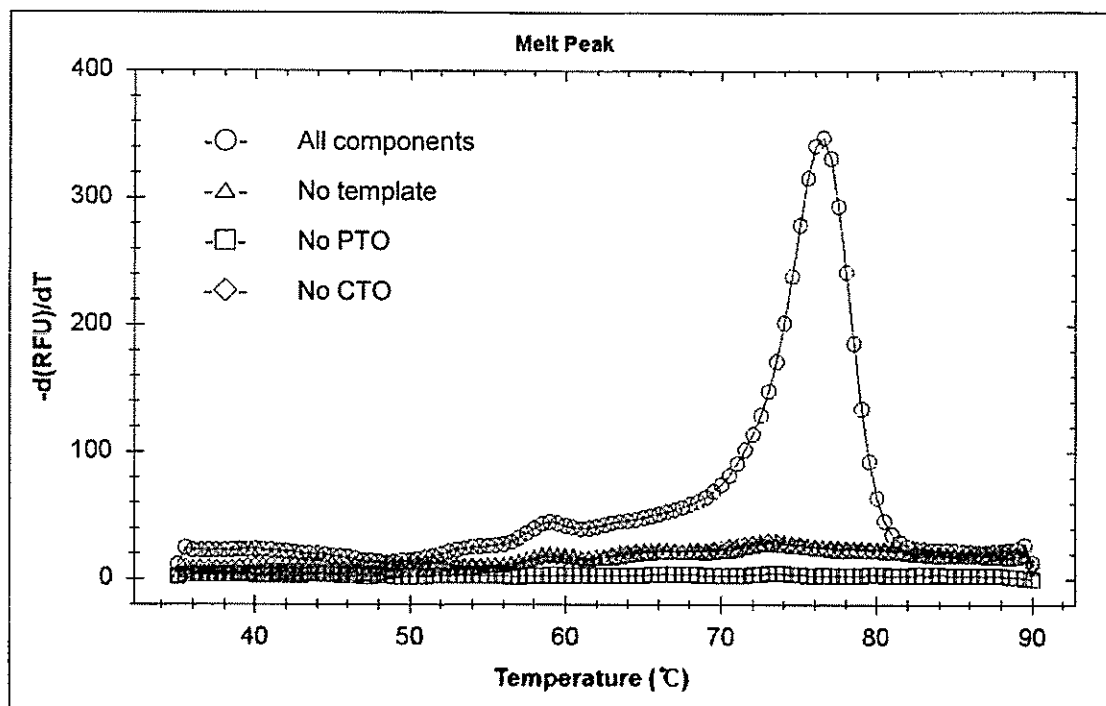
D. Extension of PTO fragment



E. Detection at a pre-determined temperature



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Fig. 14

Template ¹⁾	Upstream primer	PTO ²⁾	CTO ³⁾	T _m (°C)	
				Target ⁴⁾	Non-target ⁵⁾
+	+	+	+	76.5	-
-	+	+	+	-	-
+	+	-	+	-	-
+	+	+	-	-	-

¹⁾ Template is a synthetic oligonucleotide for *Neisseria gonorrhoeae* gene.

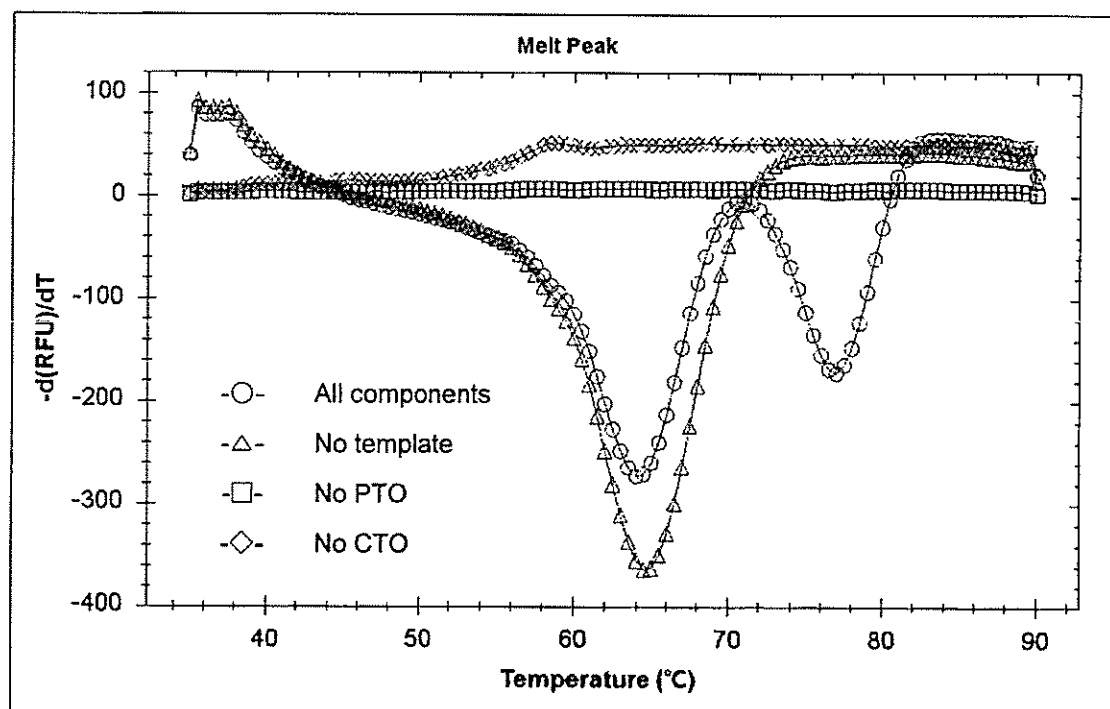
²⁾ PTO has no label.

³⁾ CTO has a quencher molecule and a fluorescent reporter molecule in the templating portion.

⁴⁾ Target represents a target signal which is T_m of the extended duplex.

⁵⁾ Non-target represents a non-target signal which is T_m of the hybrid between uncleaved PTO and CTO.

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Fig. 15

Template ¹⁾	Upstream primer	PTO ²⁾	CTO ³⁾	T _m (°C)	
				Target ⁴⁾	Non-target ⁵⁾
+	+	+	+	77.0	64.0
-	+	+	+	-	64.5
+	+	-	+	-	-
+	+	+	-	-	-

¹⁾ Template is a synthetic oligonucleotide for *Neisseria gonorrhoeae* gene.

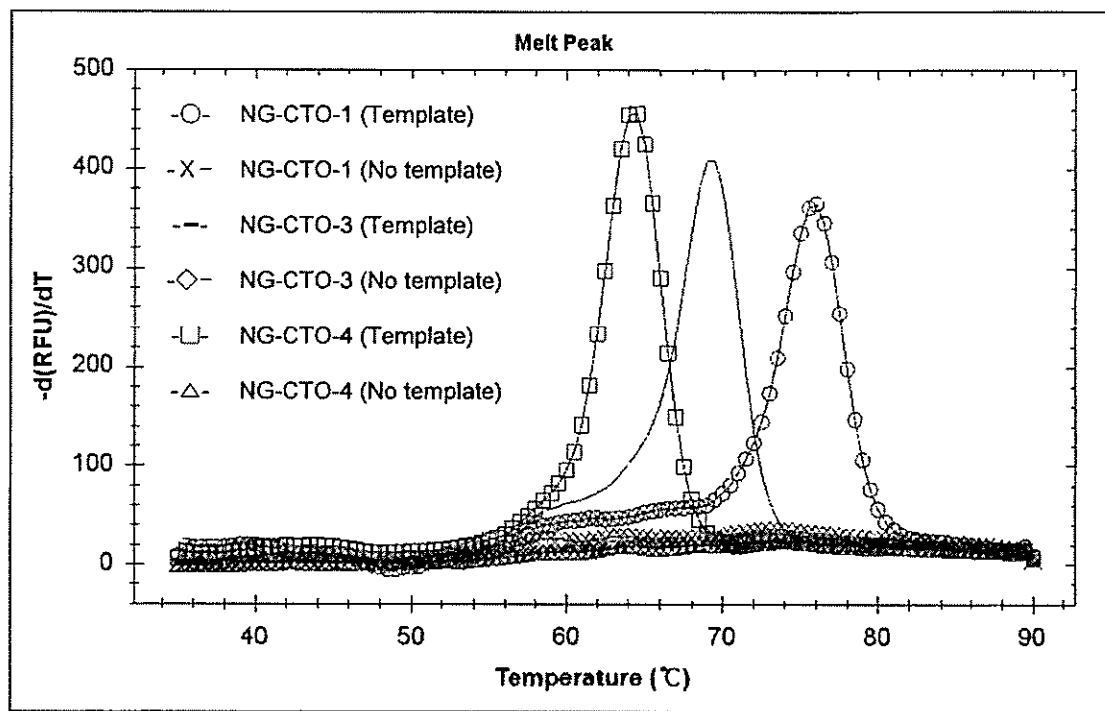
²⁾ PTO has a quencher molecular at its 5'-end.

³⁾ CTO has a fluorescent reporter molecule at its 3'-end.

⁴⁾ Target represents a target signal which is T_m of the extended duplex.

⁵⁾ Non-target represents a non-target signal which is T_m of the hybrid between uncleaved PTO and CTO.

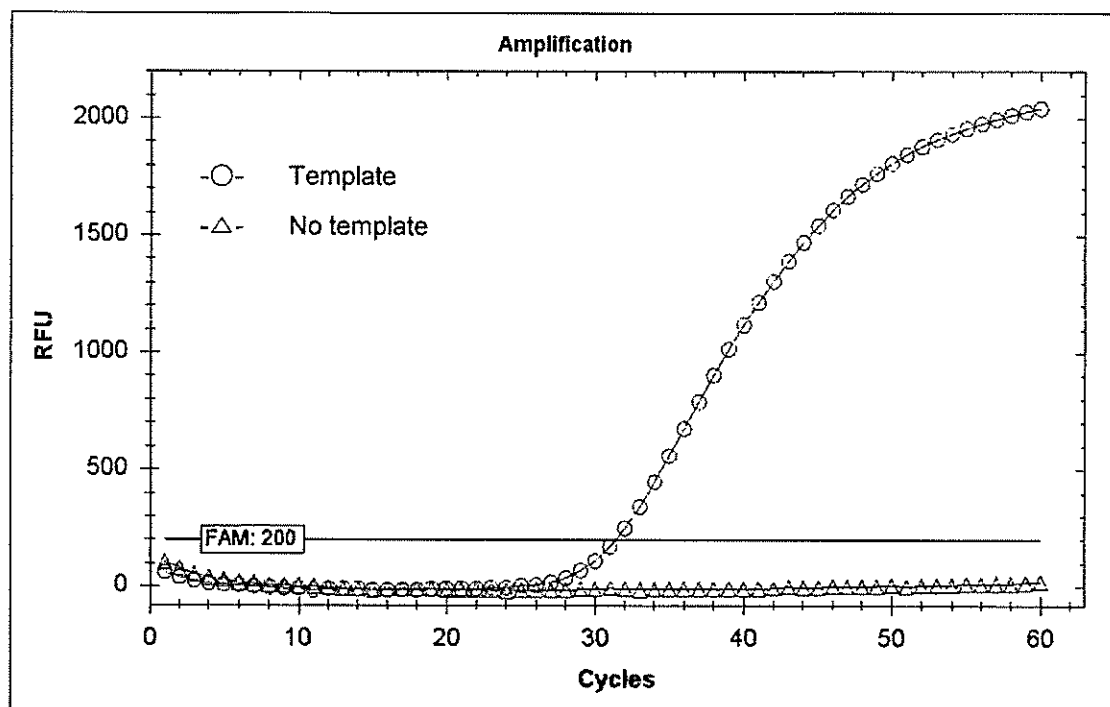
16/30

Fig. 16

Template ¹⁾	Upstream primer	PTO ²⁾	CTO ³⁾	T _m (°C) ⁴⁾
+	+	+	NG-CTO-1	76.0
-	+	+	NG-CTO-1	-
+	+	+	NG-CTO-3	69.0
-	+	+	NG-CTO-3	-
+	+	+	NG-CTO-4	64.5
-	+	+	NG-CTO-4	-

- 1) Template is a synthetic oligonucleotide for *Neisseria gonorrhoeae* gene.
- 2) PTO has no label.
- 3) CTO has a quencher molecule and a fluorescent reporter molecule in the templating portion. Three CTOs have three different sequences in their templating portions.
- 4) T_m represents melting temperature of the extended duplex.

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Fig. 17A

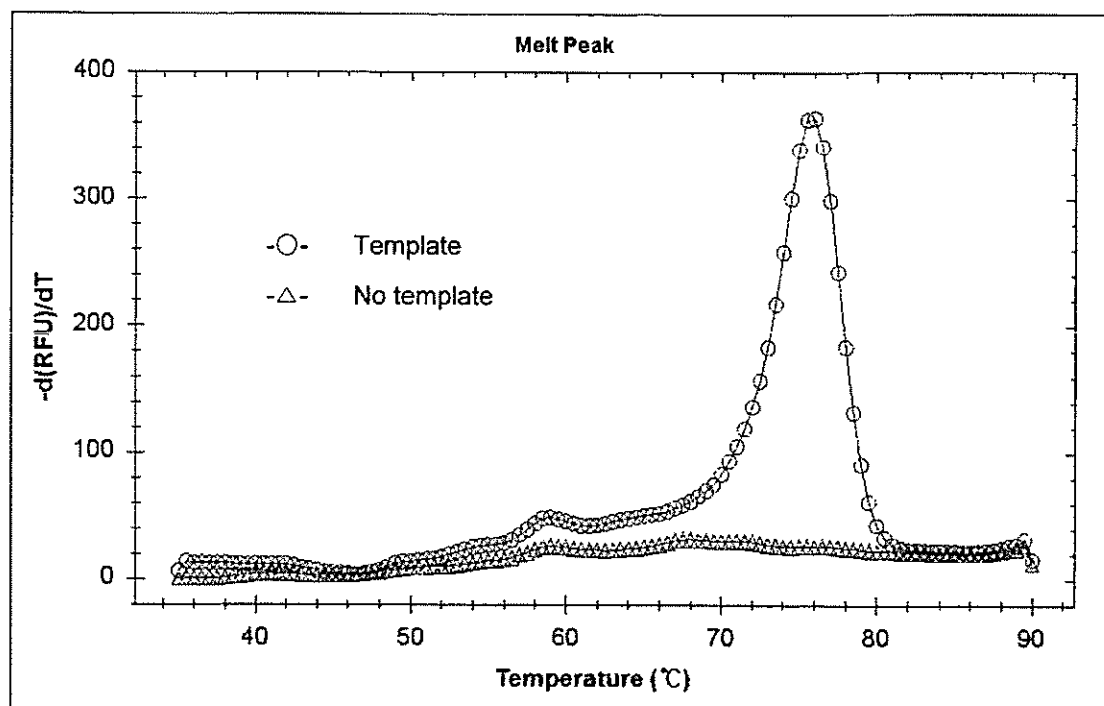
Template ¹⁾	Upstream primer	Downstream primer	PTO ²⁾	CTO ³⁾	Ct
+	+	+	+	+	31.36
-	+	+	+	+	-

¹⁾ Template is a genomic DNA of *Neisseria gonorrhoeae*.

²⁾ PTO has no label.

³⁾ CTO has a quencher molecule and a fluorescent reporter molecule in the templating portion.

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Fig. 17B

Template ¹⁾	Upstream primer	Downstream primer	PTO ²⁾	CTO ³⁾	T _m (°C) ⁴⁾
+	+	+	+	+	76.0
-	+	+	+	+	-

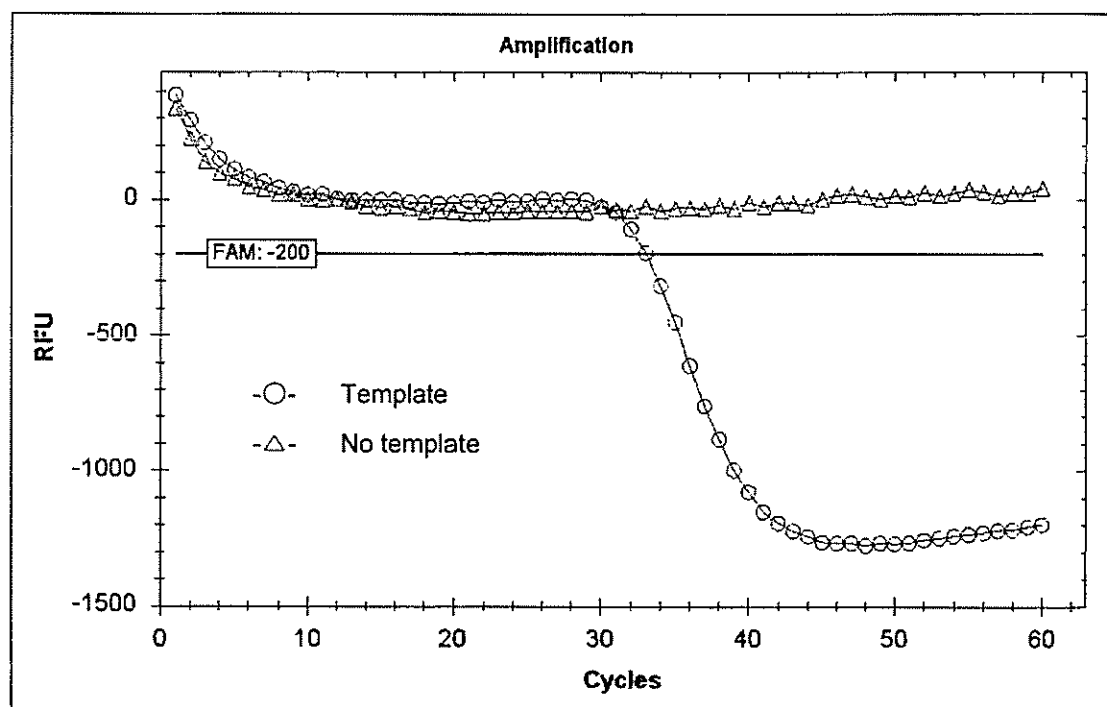
¹⁾ Template is a genomic DNA of *Neisseria gonorrhoeae*.

²⁾ PTO has no label.

³⁾ CTO has a quencher molecule and a fluorescent reporter molecule in the templating portion.

⁴⁾ T_m represents melting temperature of the extended duplex.

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Fig. 18A

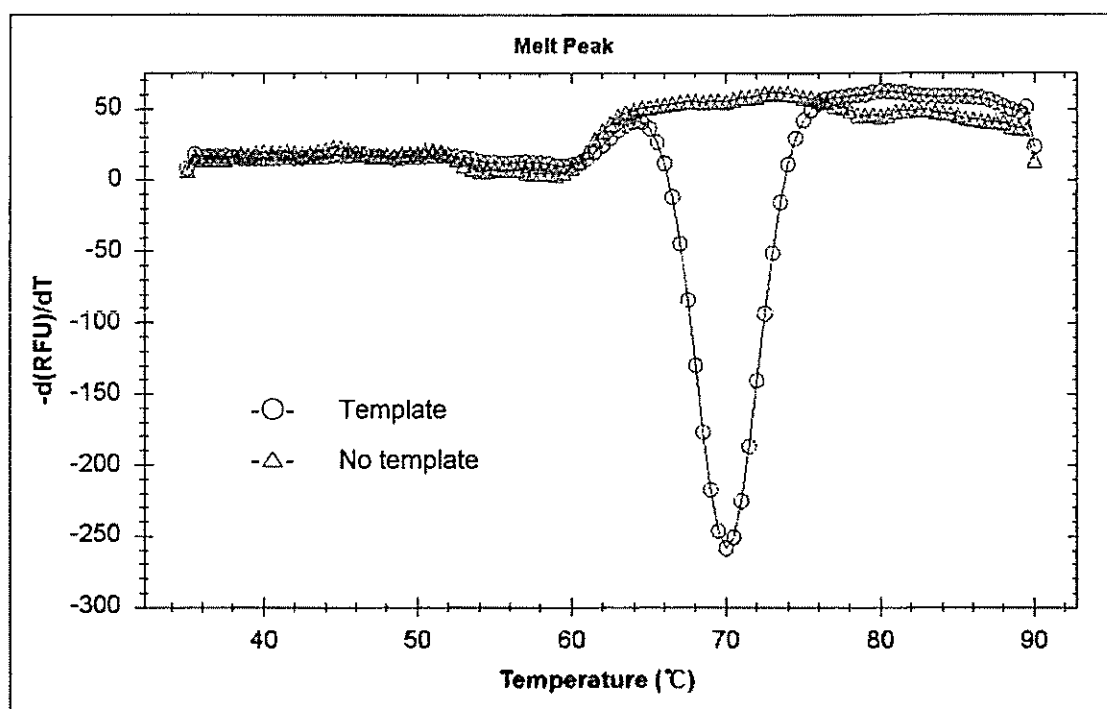
Template ¹⁾	Upstream Primer	Downstream primer	PTO ²⁾	CTO ³⁾	Ct
+	+	+	+	+	33.03
-	+	+	+	+	-

¹⁾ Template is a genomic DNA of *Neisseria gonorrhoeae*.

²⁾ PTO has no label.

³⁾ CTO has a non-natural nucleotide (iso-dC) labeled with a fluorescence reporter molecule at its 5'-end.

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Fig. 18B

Template ¹⁾	Upstream Primer	Downstream primer	PTO ²⁾	CTO ³⁾	T _m (°C) ⁴⁾
+	+	+	+	+	70.0
-	+	+	+	+	-

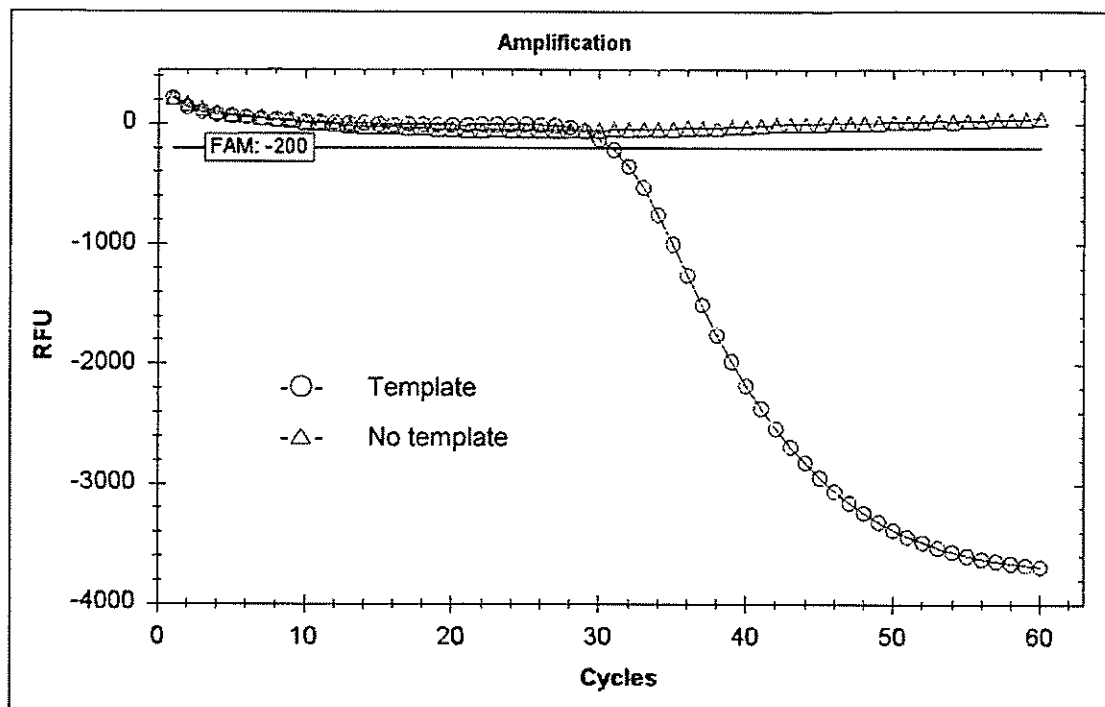
¹⁾ Template is a genomic DNA of *Neisseria gonorrhoeae*.

²⁾ PTO has no label.

³⁾ CTO has a non-natural nucleotide (iso-dC) and a fluorescence reporter molecule at its 5'-end.

⁴⁾ T_m represents melting temperature of the extended duplex.

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Fig. 19A

Template ¹⁾	Upstream primer	Downstream primer	PTO ²⁾	CTO ³⁾	Ct
+	+	+	+	+	29.79
-	+	+	+	+	-

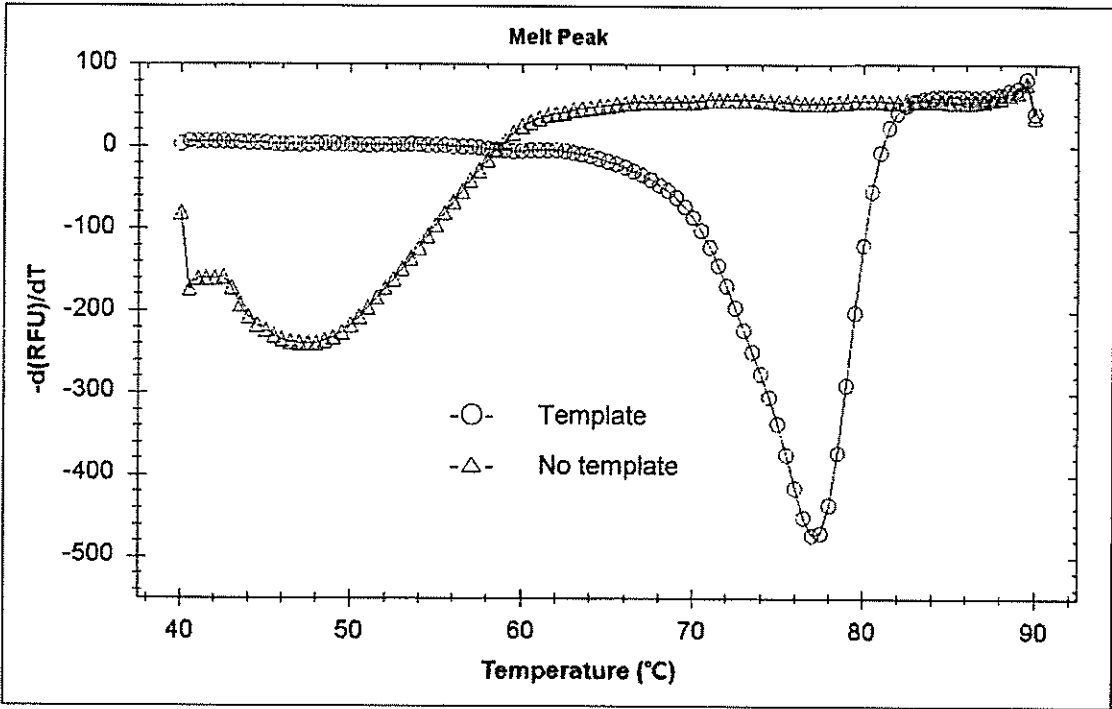
¹⁾ Template is a genomic DNA of *Neisseria gonorrhoeae*.

²⁾ PTO has a quencher molecular at its 5'-end.

³⁾ CTO has a fluorescent reporter molecule at its 3'-end.

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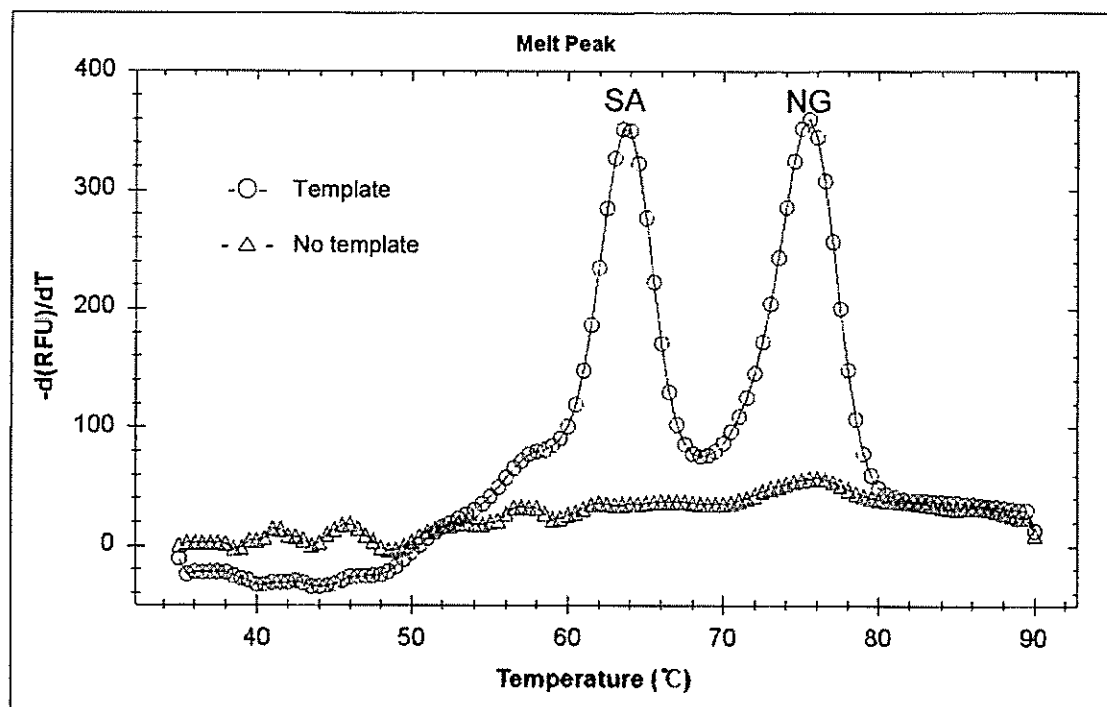
Fig. 19B



Template ¹⁾	Upstream primer	Downstream primer	PTO ²⁾	CTO ³⁾	T _m (°C)	
					Target ⁴⁾	Non-target ⁵⁾
+	+	+	+	+	76.5	-
-	+	+	+	+	-	48.0

- ¹⁾ Template is a genomic DNA of *Neisseria gonorrhoeae*.
- ²⁾ PTO has a quencher molecular at its 5'- end.
- ³⁾ CTO has a fluorescent reporter molecule at its 3'-end.
- ⁴⁾ Target represents a target signal which is T_m of the extended duplex.
- ⁵⁾ Non-target represents a non-target signal which is T_m of the hybrid between uncleaved PTO and CTO .

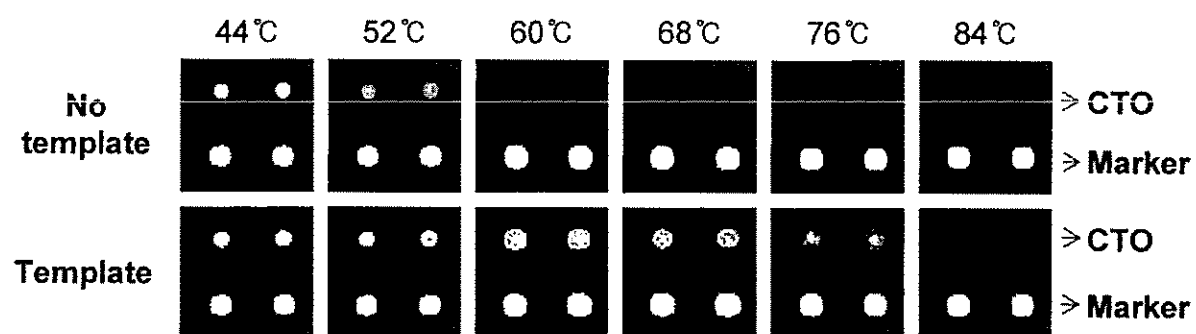
23/30

Fig. 20

Templates ¹⁾	Upstream primer	Downstream primer	PTO ²⁾	CTO ³⁾	T _m (°C) ⁴⁾
NG and SA	+	+	+	+	75.5 and 63.5
-	+	+	+	+	-

- ¹⁾ Templates are genomic DNAs of *Neisseria gonorrhoeae* (NG) and *Staphylococcus aureus* (SA).
- ²⁾ PTO has no label.
- ³⁾ CTO has a quencher molecule and a fluorescent reporter molecule in the templating portion. The CTOs for NG and SA have the same type of a fluorescent reporter molecule (FAM).
- ⁴⁾ T_m represents melting temperature of the extended duplex.

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Fig. 21A**Fluorescent image depending temperaute on microarray**

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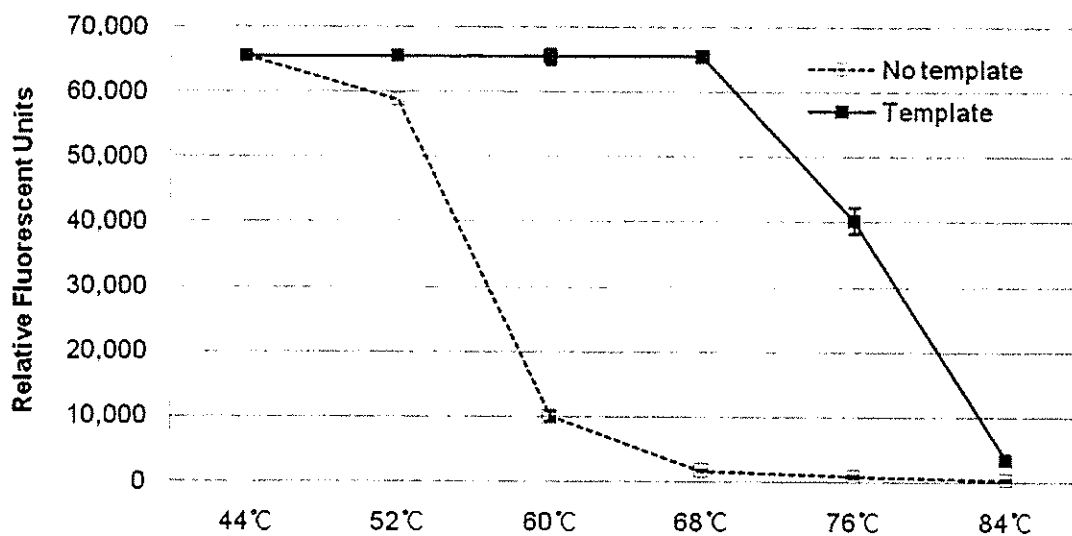
Fig. 21B**Fluorescent intensity depending on temperature on microarray**

Template ¹⁾	Upstream primer	PTO ²⁾	CTO ³⁾	Relative Fluorescent Units					
				44°C	52°C	60°C	68°C	76°C	84°C
-	+	+	+	65,438 (±1.4)	58,761 (±2,249.3)	9,989 (±473.1)	1,728 (±188.1)	862 (±184.6)	303 (±82.7)
+	+	+	+	65,434 (±3.5)	65,424 (±11.3)	65,397 (±9.9)	65,393 (±12.7)	40,184 (±3,297.9)	3,490 (±246.8)

1) Template is a synthetic oligonucleotide for *Neisseria gonorrhoeae* gene.

2) PTO has a fluorescent reporter molecule at its 5'-end.

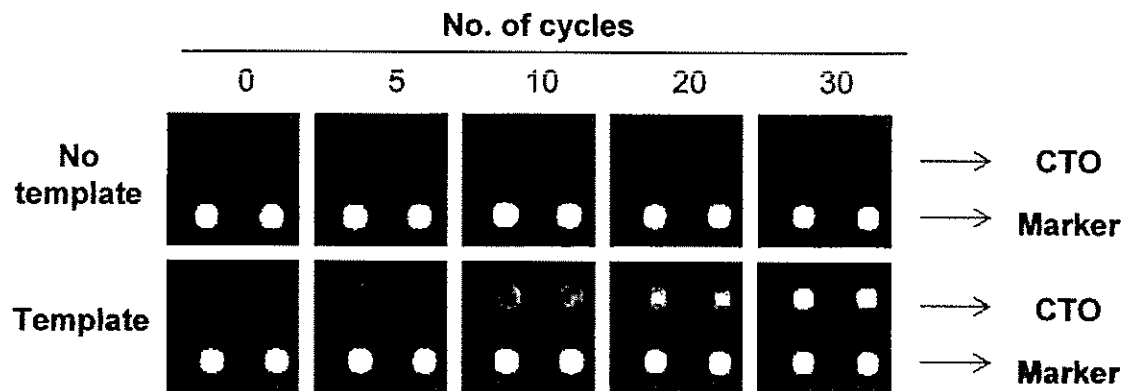
3) CTO is immobilized on the surface of solid substrate by using an amino group at its 5'-end.



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Fig. 22A

**Fluorescent images depending on cycle numbers on
microarray**



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Fig. 22B

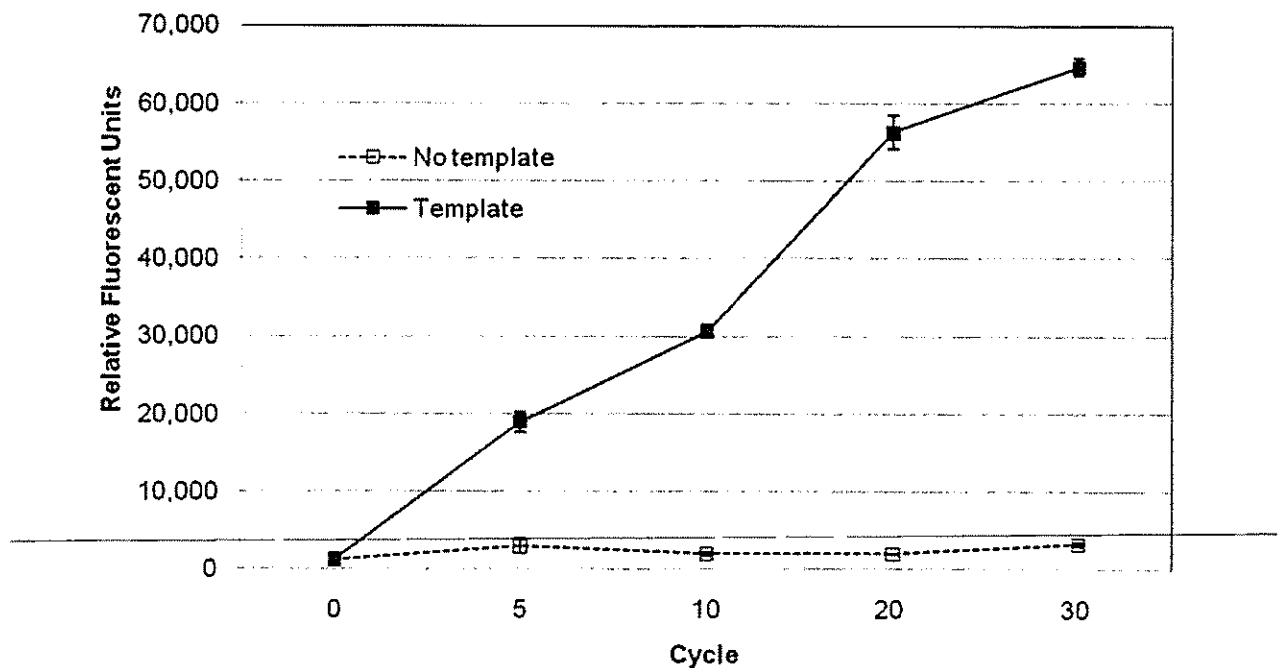
Change of fluorescence intensity depending on cycle numbers on microarray

Template ¹⁾	Upstream primer	PTO ²⁾	CTO ³⁾	Relative Fluorescent Units					
				No. of cycles :	0	5	10	20	30
-	+	+	+		1,216 (±86.3)	3,012 (±929.1)	1,988 (±276.5)	1,961 (±25.5)	3,261 (±279.3)
+	+	+	+		1,304 (±0.7)	18,939 (±1,342.1)	30,619 (±285.0)	56,248 (±2,208.3)	64,645 (±1,110.2)

1) Template is a synthetic oligonucleotide for *Neisseria gonorrhoeae* gene.

2) PTO has a fluorescent reporter molecule at its 5'-end.

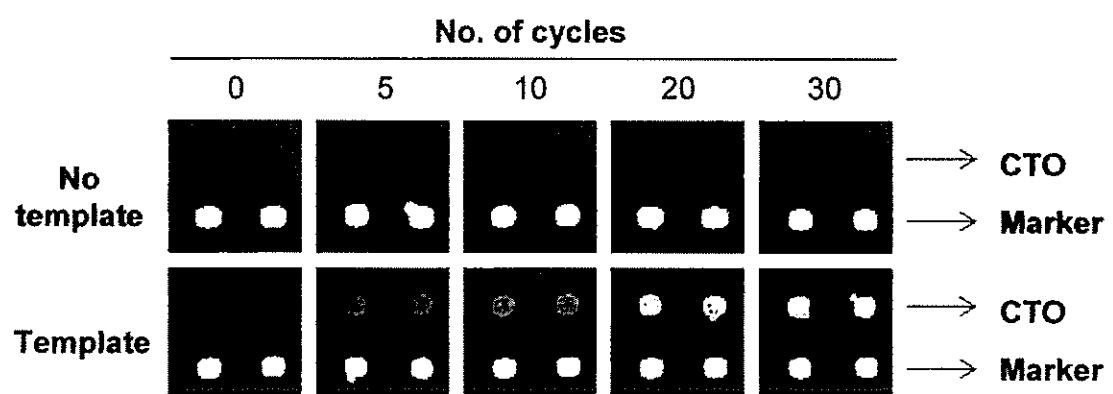
3) CTO is immobilized on the surface of solid substrate by using an amino group at its 5'-end.



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Fig. 23A

**Fluorescent images depending on cycle numbers on
microarray**



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Fig. 23B

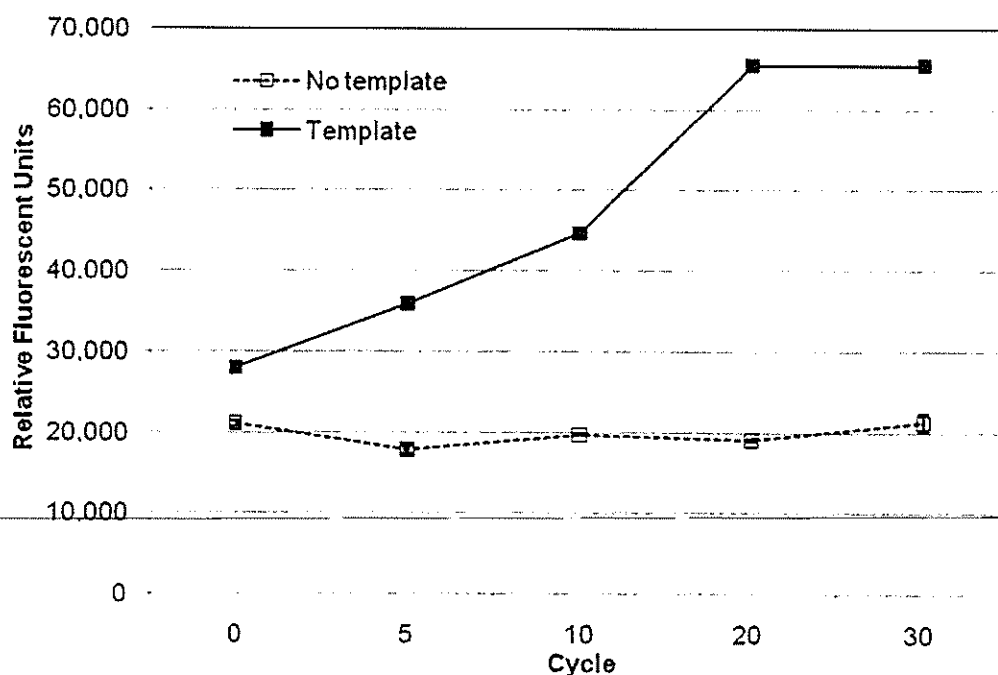
Change of fluorescence intensity depending on cycle numbers on microarray

Template ¹⁾	Upstream primer	PTO ²⁾	CTO ³⁾	Relative Fluorescent Units					
				No. of cycles :	0	5	10	20	30
-	+	+	+		21,047 (±334.5)	17,867 (±478.7)	19,739 (±82.0)	19,144 (±176.8)	21,259 (±1,130.7)
+	+	+	+		28,078 (±460.3)	35,967 (±555.1)	44,674 (±186.0)	65,423 (±2.1)	65,426 (±2.8)

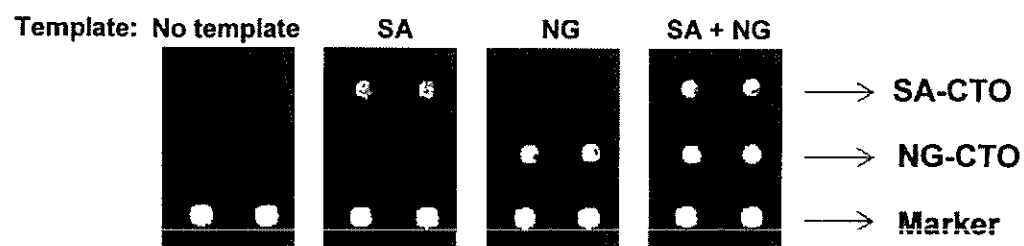
1) Template is a synthetic oligonucleotide for *Neisseria gonorrhoeae* gene.

2) PTO has no label.

3) CTO has a quencher molecule and a fluorescent reporter molecule in the templating portion. CTO is immobilized on the surface of solid substrate by using an amino group at its 3'-end.



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Fig. 24**Fluorescent image on microarray****Fluorescent intensity on microarray**

Template ¹⁾	Primers ²⁾	PTO ³⁾	CTO ⁴⁾	Relative Fluorescent Units	
				SA	NG
-	+	+	+	448 (± 29.7)	2,522 (± 116.0)
SA	+	+	+	65,192 (± 198.7)	4,183 (± 323.9)
NG	+	+	+	603 (± 36.8)	65,332 (± 1.4)
SA and NG	+	+	+	65,302 (± 0.7)	65,302 (± 0.7)

¹⁾ Template is a genomic DNA of *Staphylococcus aureus* (SA) or/and a genomic DNA of *Neisseria gonorrhoeae* (NG).

²⁾ Primers are a upstream and a downstream primer for PCR.

³⁾ PTO has a fluorescent reporter molecule at its 5'-end.

⁴⁾ CTO is immobilized on the surface of solid substrate by using an amino group at its 5'-end.