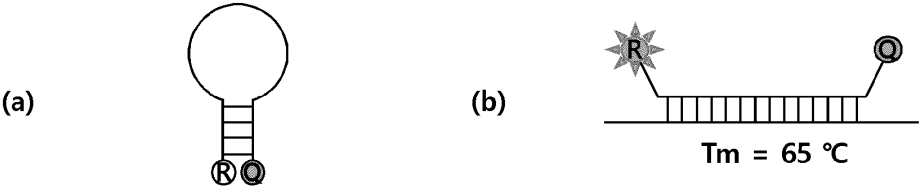


[Fig. 1a]

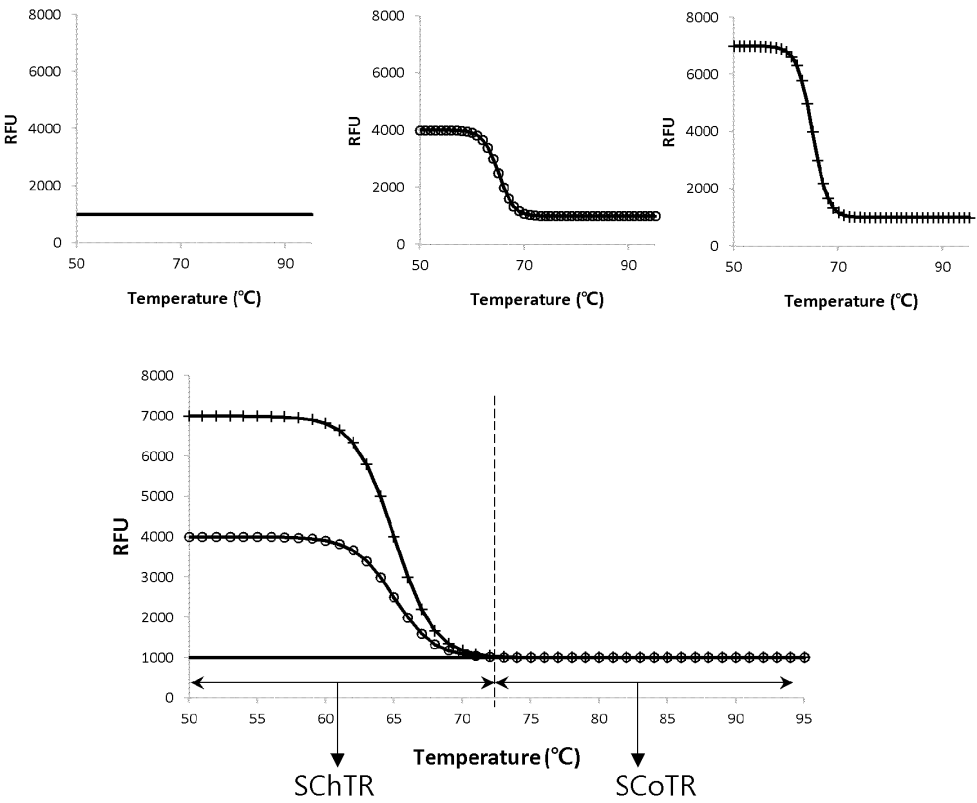
(1) Molecular beacon



(2) Melt curve

The ratio (%) of the amounts of (a) to (b)

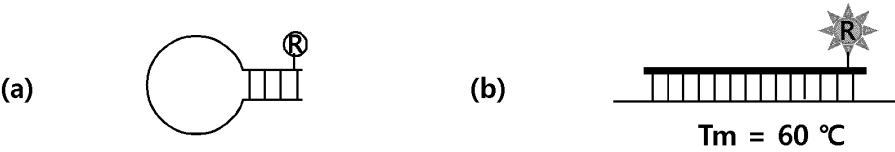
	Initial cycle	Intermediate cycle	Late cycle
(a)	100	50	0
(b)	0	50	100



Ⓡ : Reporter dye Ⓞ : Quencher

[Fig. 1b]

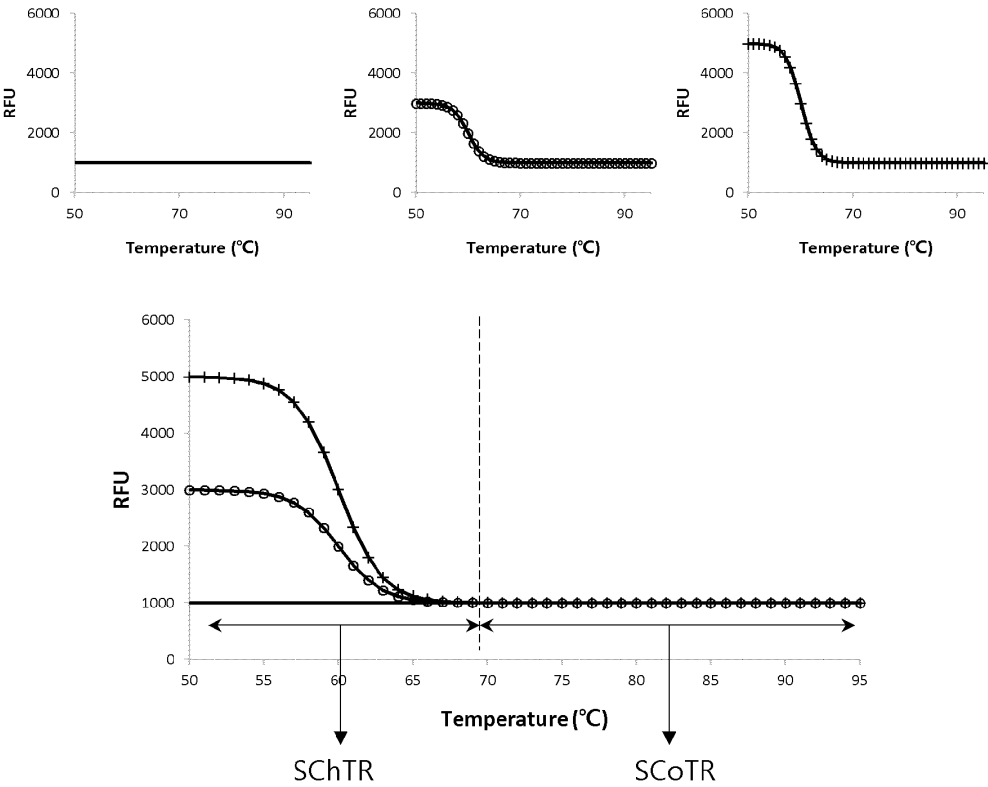
(1) Lux probe



(2) Melt curve

The ratio (%) of the amounts of (a) to (b)

	Initial cycle	Intermediate cycle	Late cycle
(a)	100	50	0
(b)	0	50	100



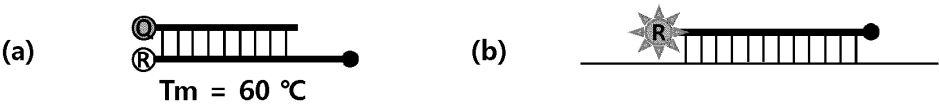
Ⓡ : Reporter dye ● : Spacer C3

(1) Hybrid probe

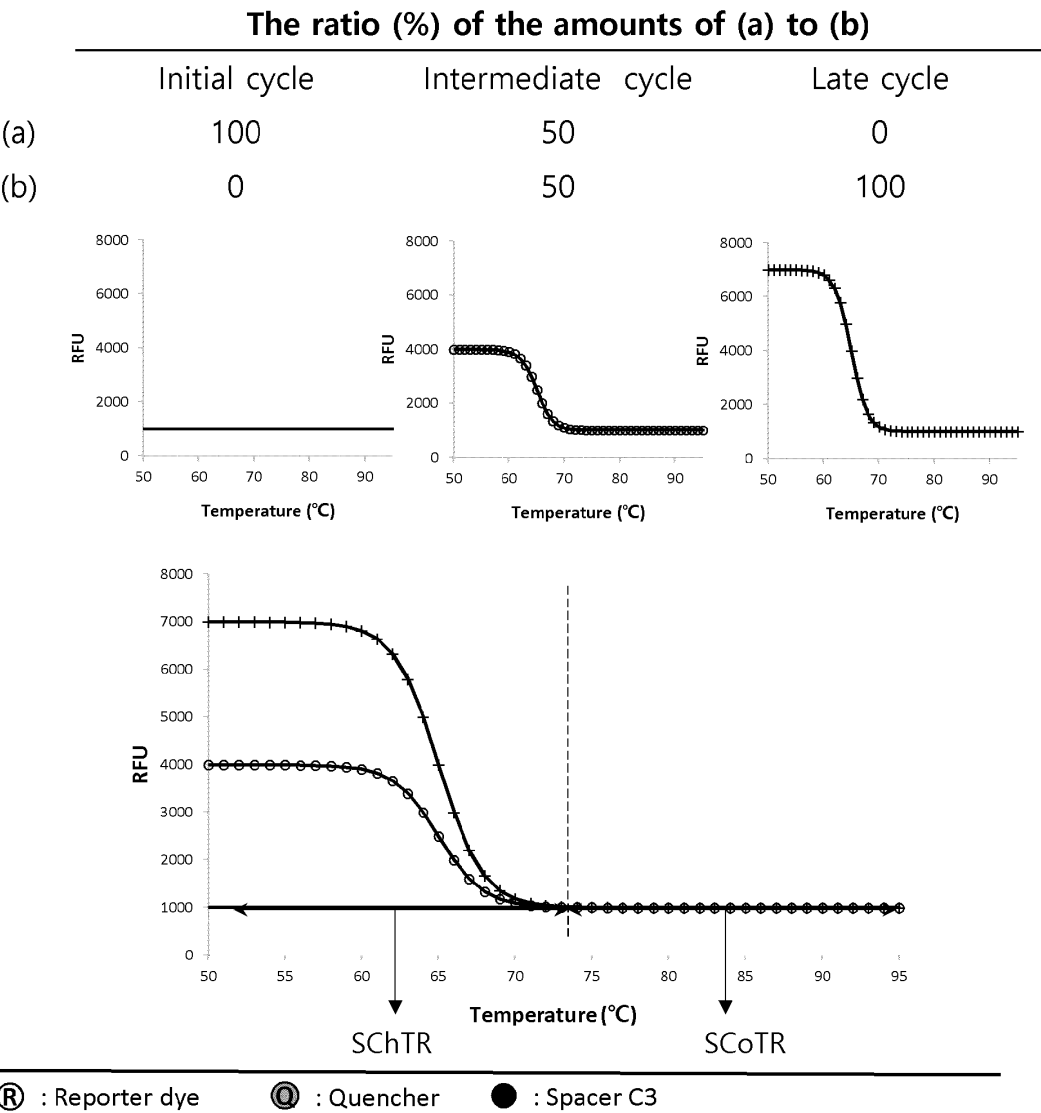


Ⓡ : Reporter dye Ⓢ : Quencher ● : Spacer C3

[Fig. 1d]
(1) Yin-Yang probe

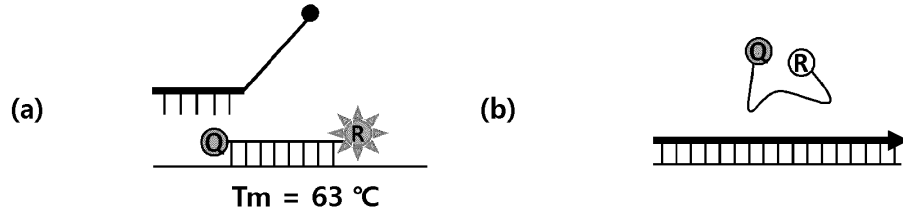


(c) Melt curve



[Fig. 1e]

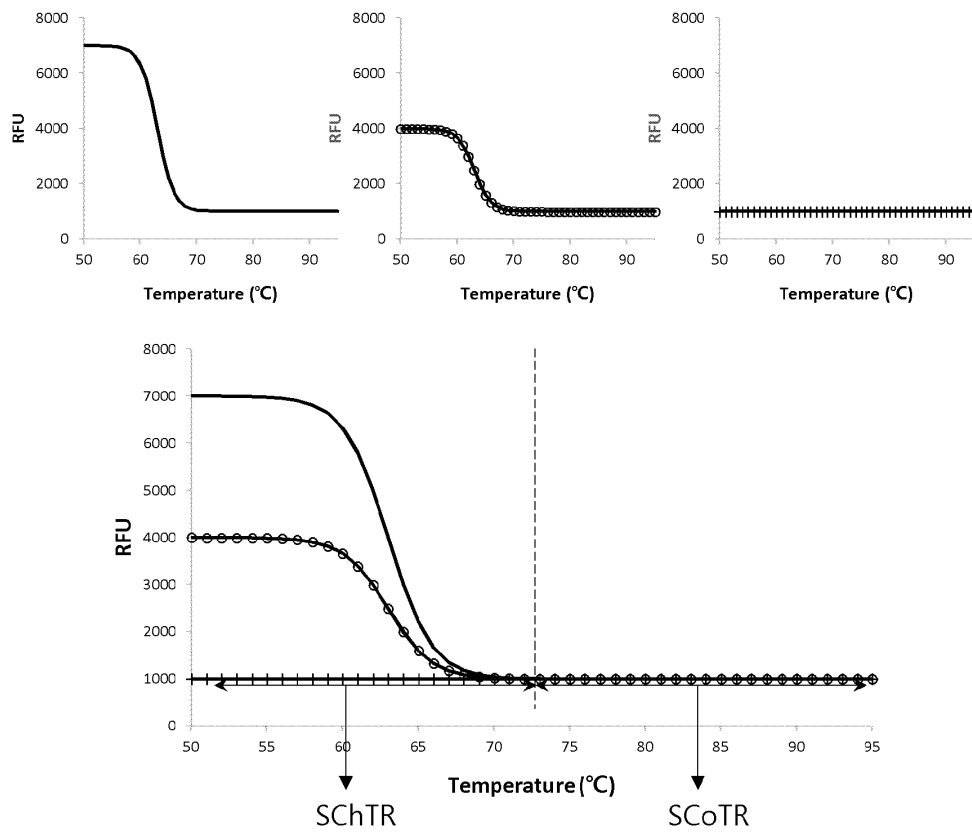
(1) PTOCE-based Method : PCE-NH



(c) Melt curve

The ratio (%) of the amounts of (a) to (b)

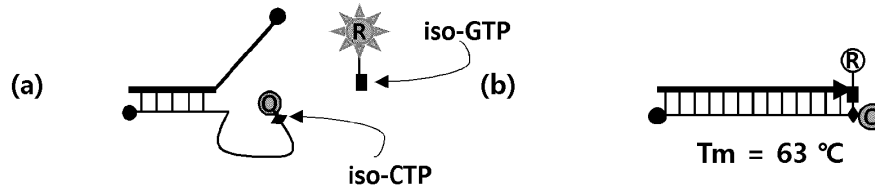
	Initial cycle	Intermediate cycle	Late cycle
(a)	100	50	0
(b)	0	50	100



® : Reporter dye ® : Quencher ● : Spacer C3

[Fig. 1f]

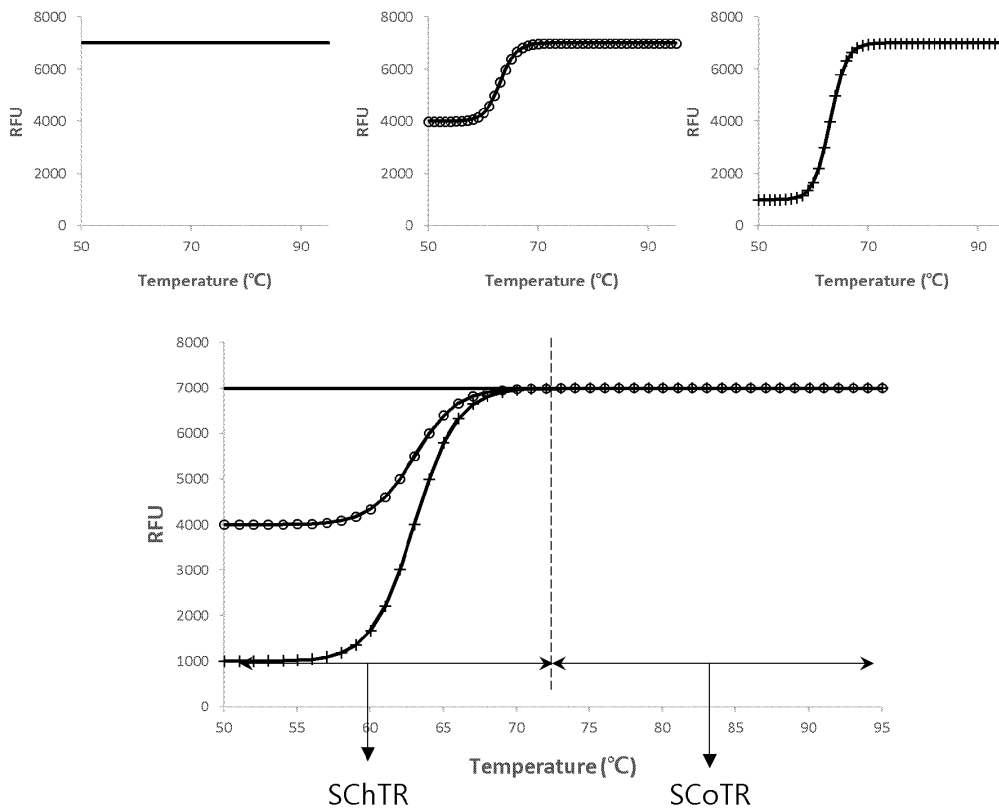
(1) PTOCE-based Method : Incorporation label



(c) Melt curve

The ratio (%) of the amounts of (a) to (b)

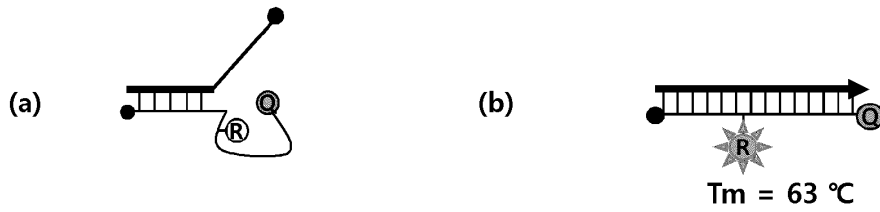
	Initial cycle	Intermediate cycle	Late cycle
(a)	100	50	0
(b)	0	50	100



® : Reporter dye ② : Quencher ● : Spacer C3

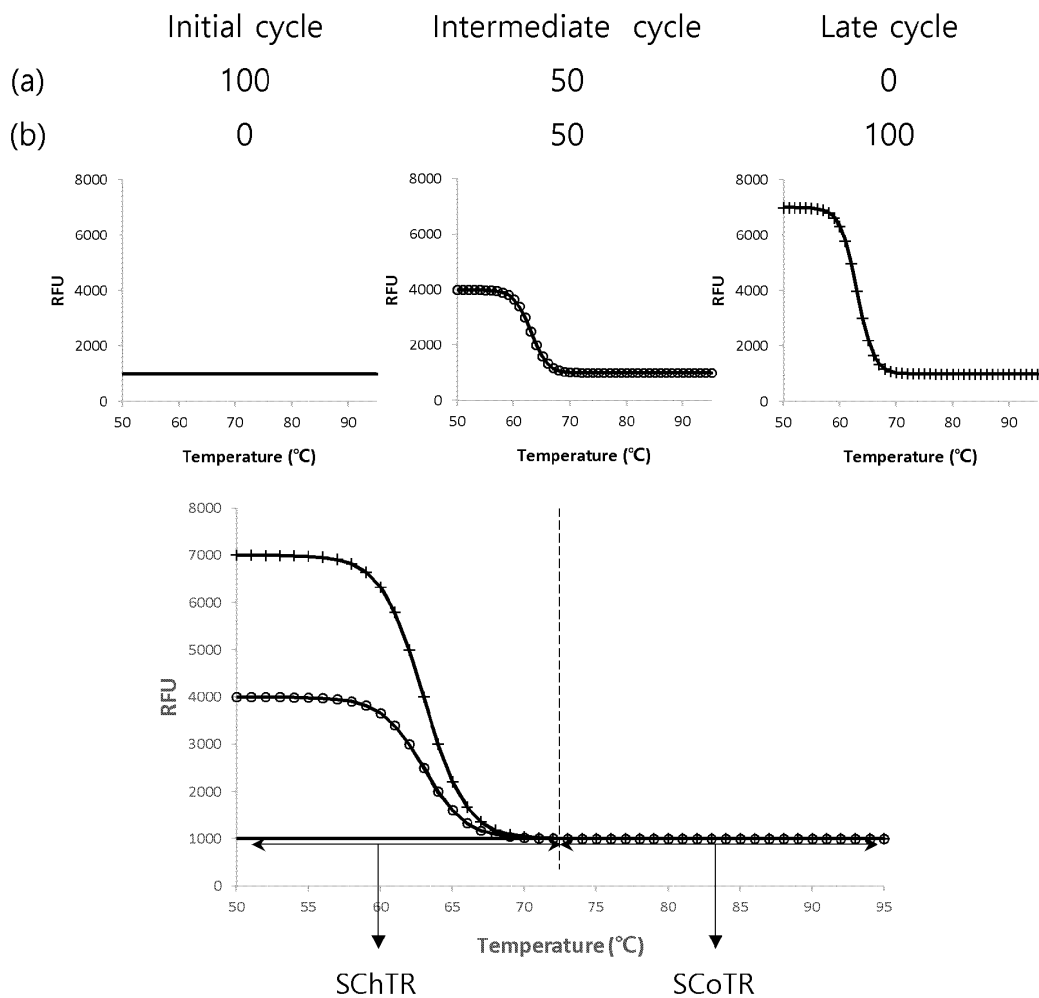
[Fig. 1g]

(1) PTOCE-based Method : Fluorescent and non-fluorescent labels



(c) Melt curve

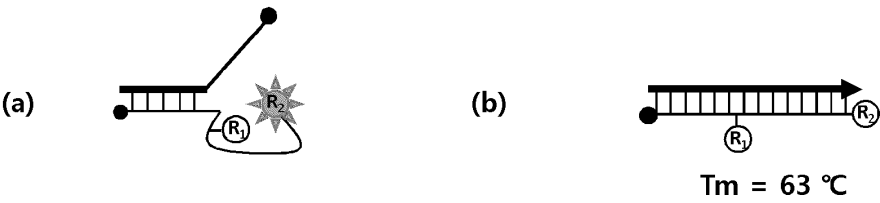
The ratio (%) of the amounts of (a) to (b)



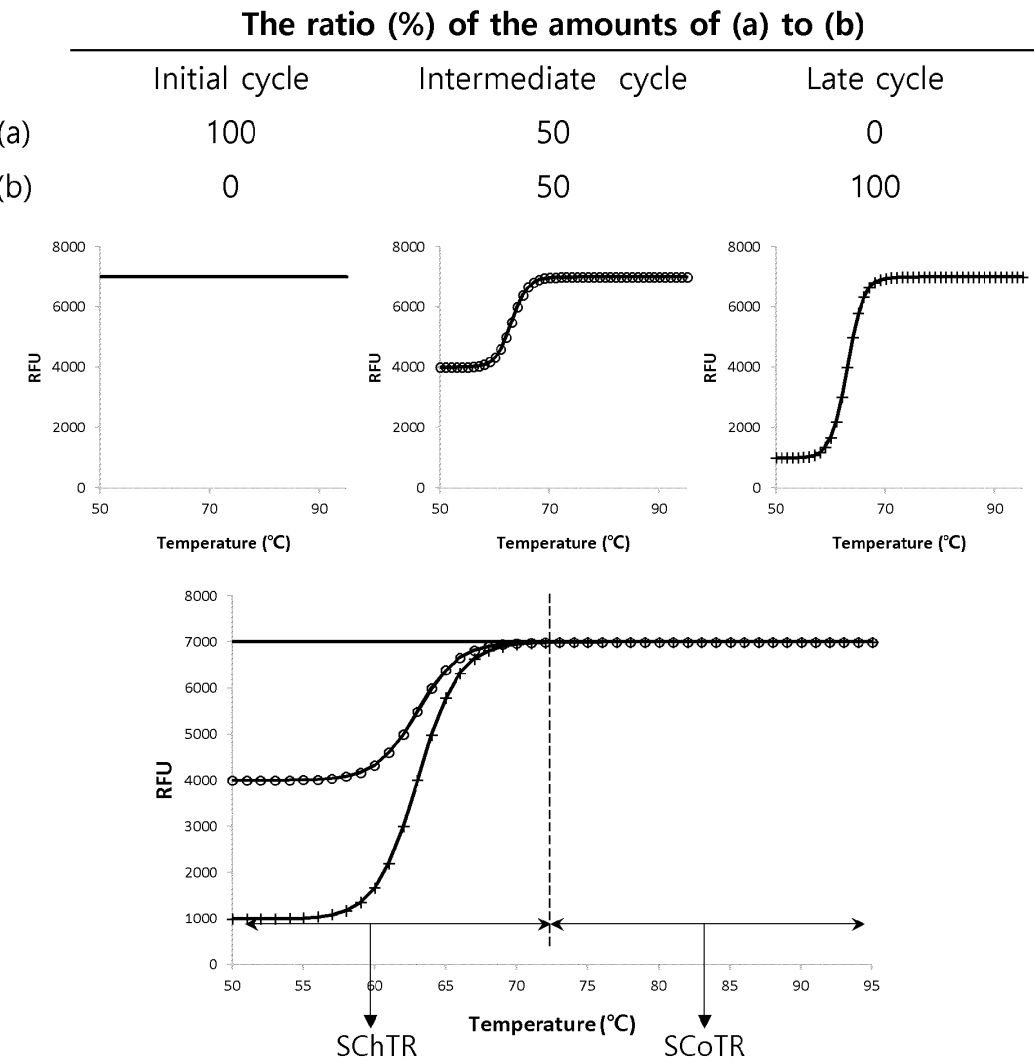
® : Reporter dye ® : Quencher ● : Spacer C3

[Fig. 1h]

(1) PTOCE-based Method : Fluorescent and fluorescent labels



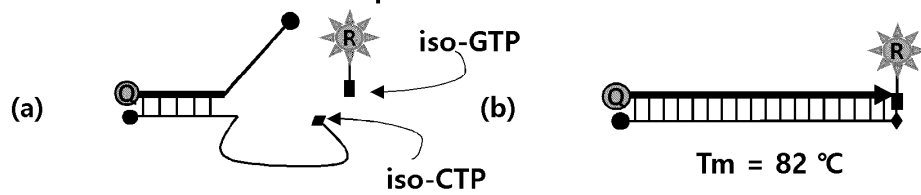
(2) Melt curve



Ⓡ : Reporter dye Ⓚ : Quencher ● : Spacer C3

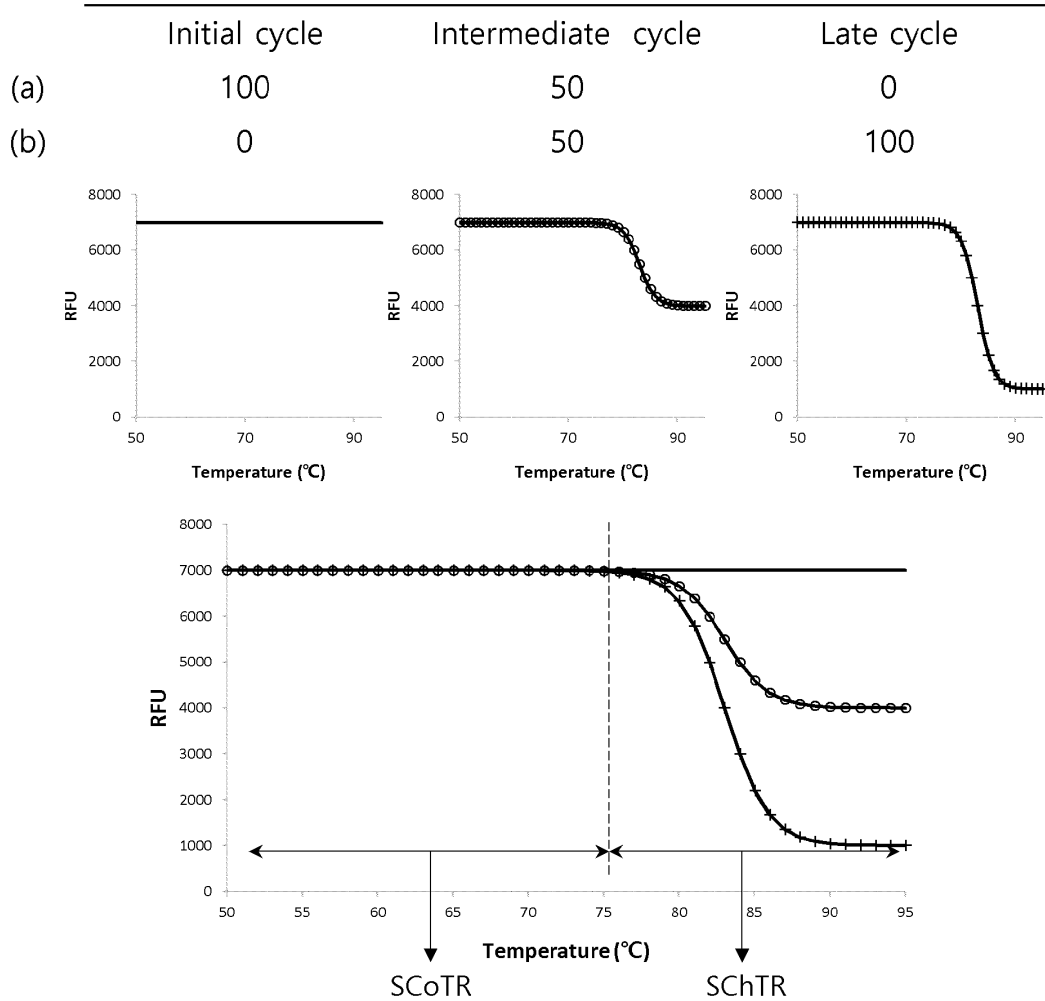
[Fig. 2a]

(1) PTOCE-based Method : Incorporation label



(c) Melt curve

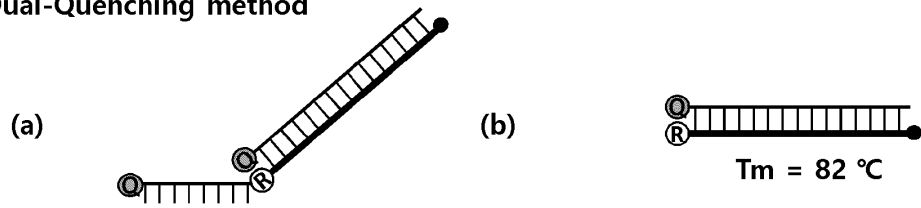
The ratio (%) of the amounts of (a) to (b)



® : Reporter dye ® : Quencher ● : Spacer C3

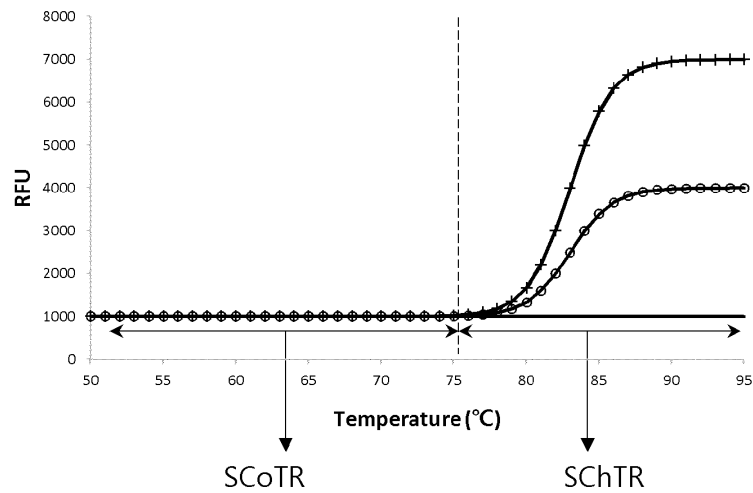
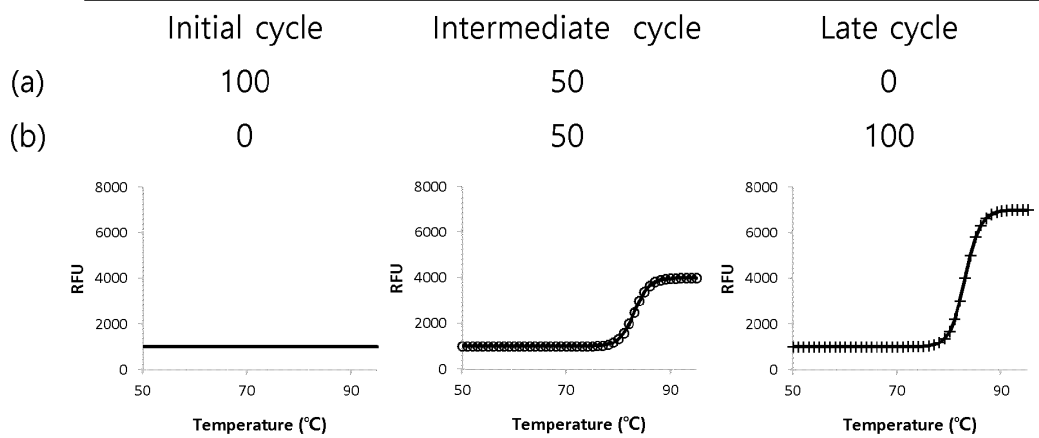
[Fig. 2b]

(1) Dual-Quenching method



(c) Melt curve

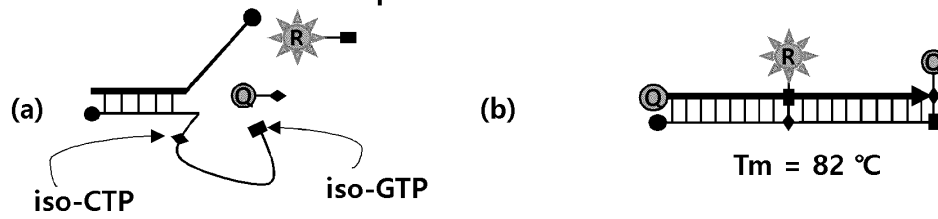
The ratio (%) of the amounts of (a) to (b)



® : Reporter dye ® : Quencher ● : Spacer C3

[Fig. 2c]

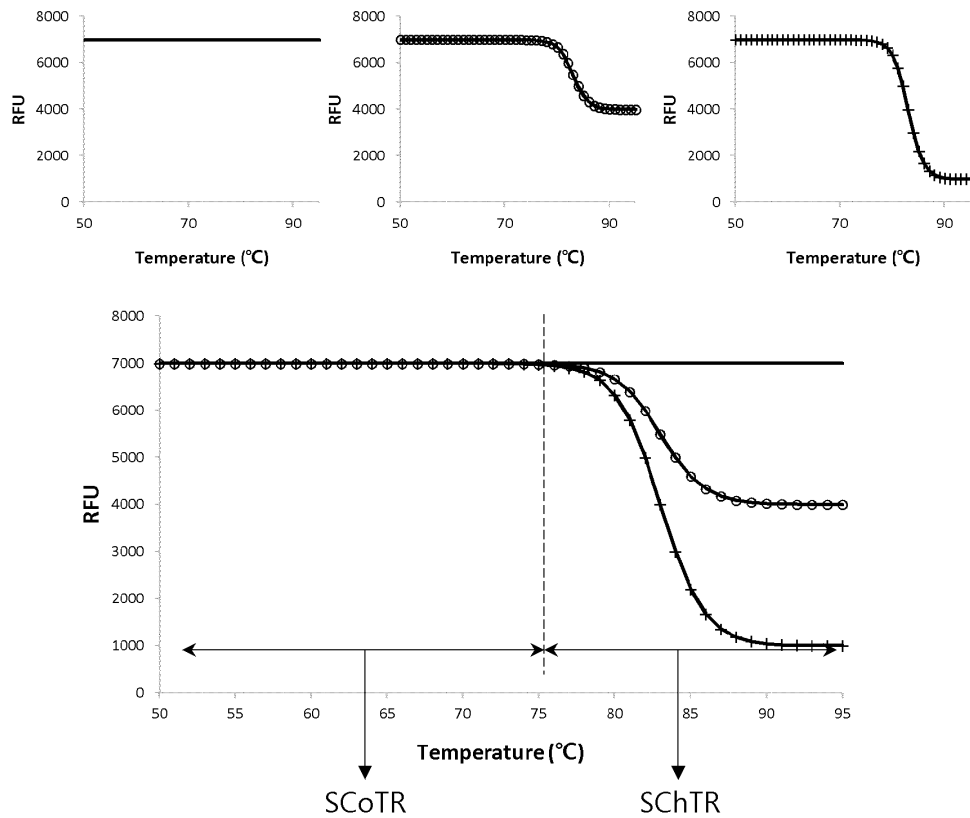
(1) PTOCE-based Method : Incorporation label



(c) Melt curve

The ratio (%) of the amounts of (a) to (b)

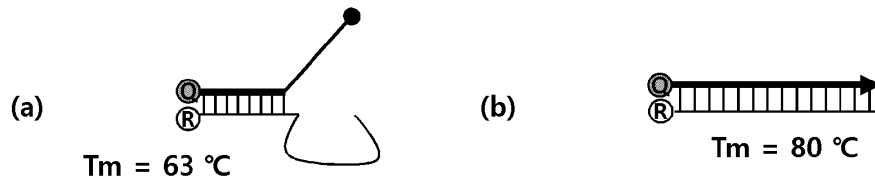
	Initial cycle	Intermediate cycle	Late cycle
(a)	100	50	0
(b)	0	50	100



® : Reporter dye Ⓚ : Quencher ● : Spacer C3

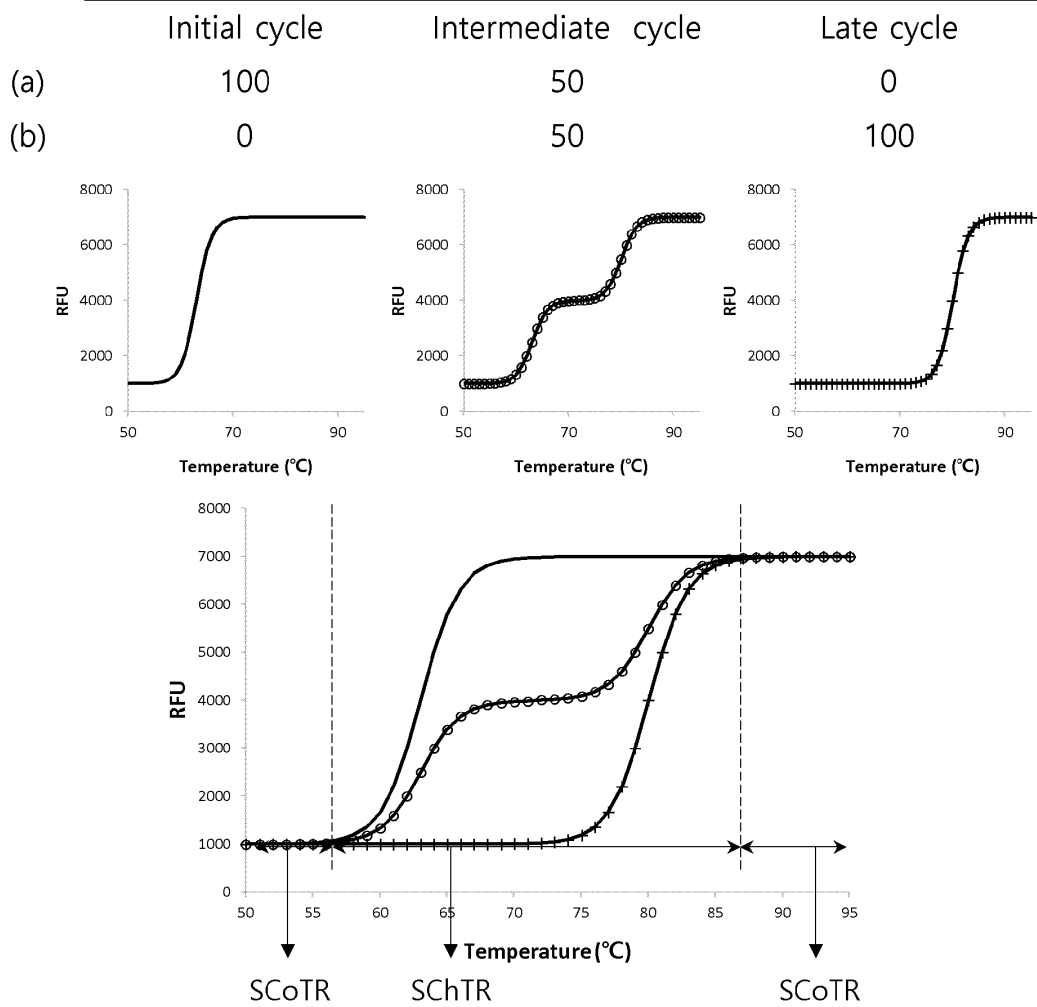
[Fig. 3a]

(1) PTOCE-based Method



(c) Melt curve

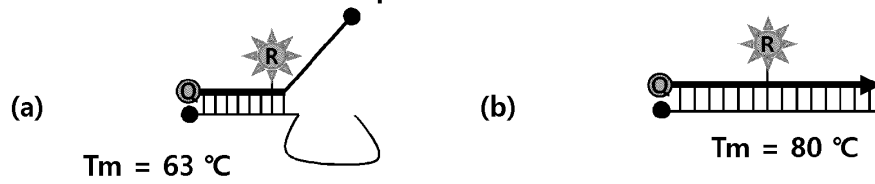
The ratio (%) of the amounts of (a) to (b)



® : Reporter dye ® : Quencher ● : Spacer C3

[Fig. 3b]

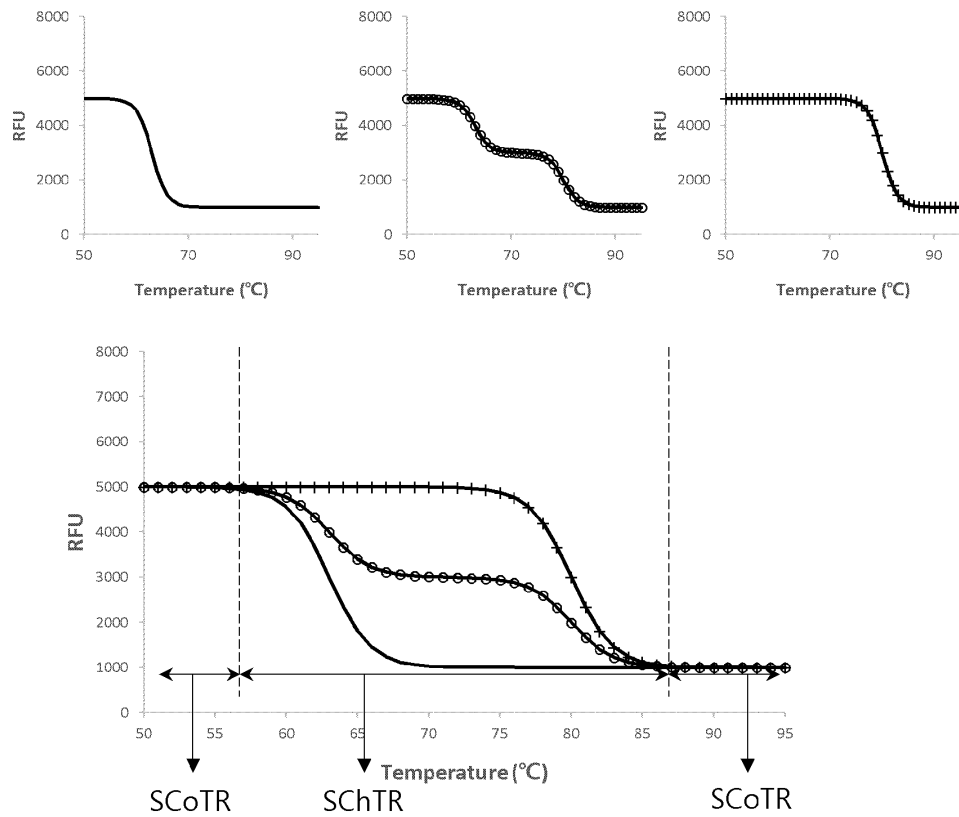
(1) PTOCE-based Method : Incorporation label



(c) Melt curve

The ratio (%) of the amounts of (a) to (b)

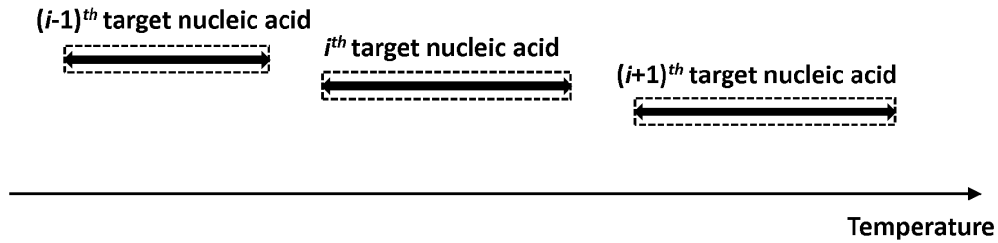
	Initial cycle	Intermediate cycle	Late cycle
(a)	100	50	0
(b)	0	50	100



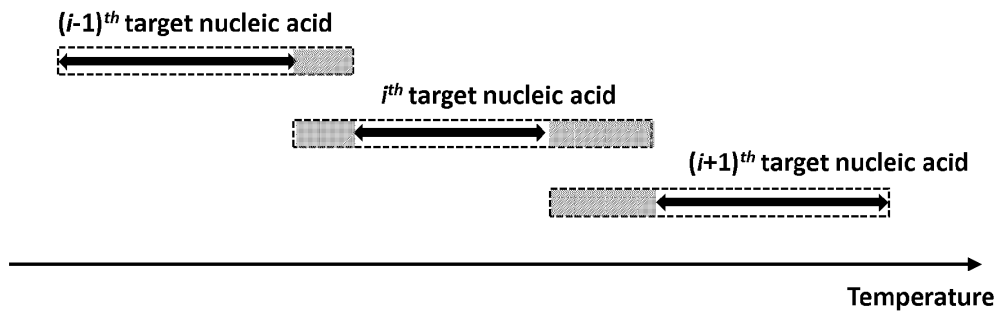
® : Reporter dye ® : Quencher ● : Spacer C3

[Fig. 4]

(a) In case that SChTRs do not overlap each other



(b) In case that SChTRs overlap partially each other



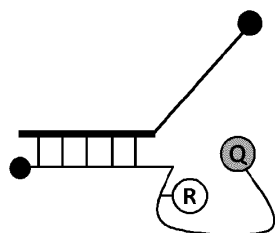
 : Signal-Changing temperature range(SChTR)

 : Overlapping range

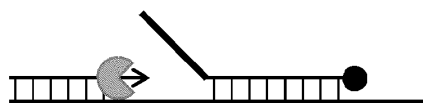
 : Selectable range for detection temperature

[Fig. 5]

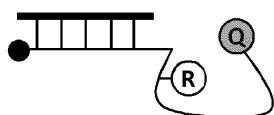
(a) The absence of a target nucleic acid or prior to incubation



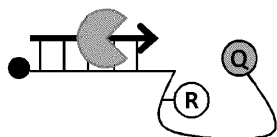
(b) Hybridization of mediation oligonucleotide with target nucleic acid and cleavage of the mediation oligonucleotide to release a fragment



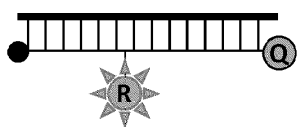
(c) Hybridization of the fragment with a capture oligonucleotide



(d) Extension of the fragment



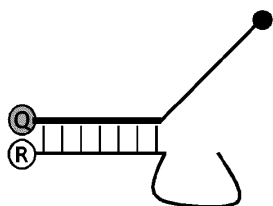
(e) Generation of an extended duplex



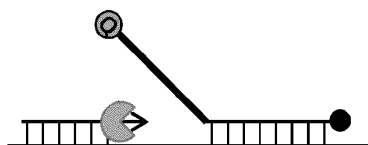
Ⓡ : Reporter dye Ⓢ : Quencher ● : Spacer C3

[Fig. 6]

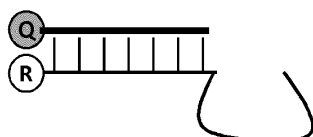
(a) The absence of a target nucleic acid or prior to incubation



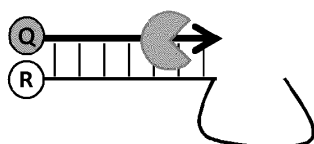
(b) Hybridization of mediation oligonucleotide with target nucleic acid and cleavage of the mediation oligonucleotide to release a fragment



(c) Hybridization of the fragment with a capture oligonucleotide



(d) Extension of the fragment



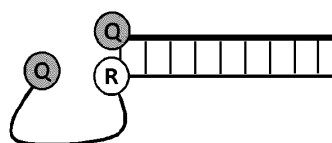
(e) Generation of an extended duplex



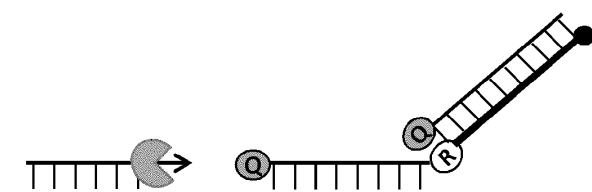
Ⓡ : Reporter dye Ⓢ : Quencher ● : Spacer C3

[Fig. 7]

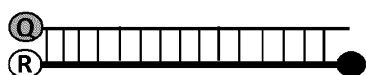
(a) The absence of a target nucleic acid or prior to incubation



(b) Hybridization of mediation oligonucleotide with target nucleic acid and cleavage of the mediation oligonucleotide to release an activated Tag Duplex fragment



(c) Association or dissociation of the activated Tag Duplex fragment



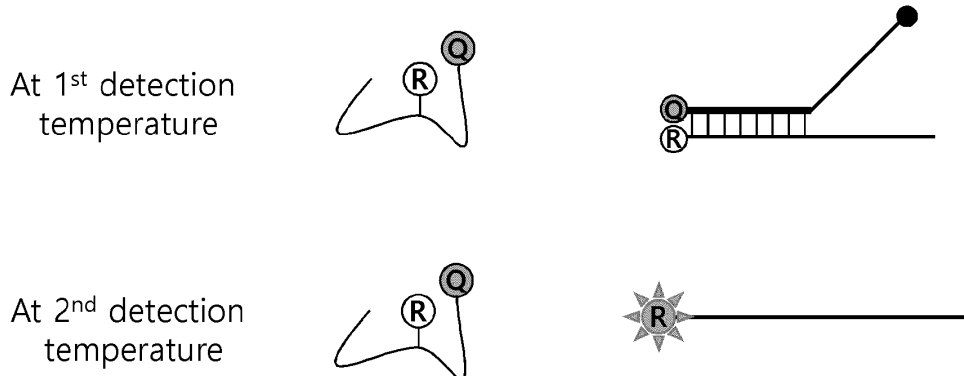
Ⓡ : Reporter dye Ⓚ : Quencher ● : Spacer C3

[Fig. 8]

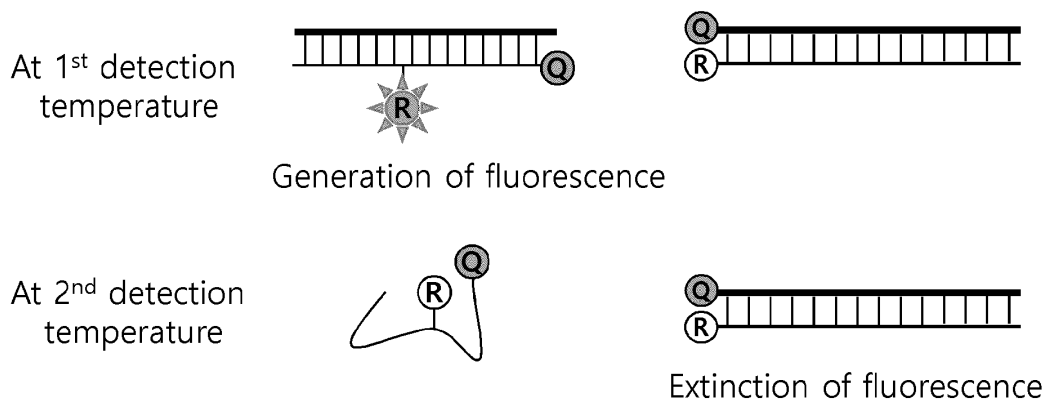
**A composition for
detecting 1st target
nucleic acid
(UnderSC)**

**A composition for
detecting 2nd target
nucleic acid
(InterSC)**

(a) In the absence of a corresponding target nucleic acid



(b) In the presence of a corresponding target nucleic acid



Ⓡ : Reporter dye Ⓢ : Quencher ● : Spacer C3

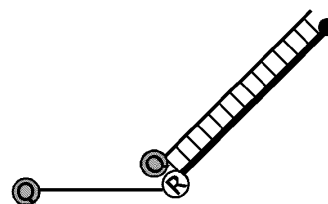
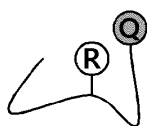
[Fig. 9]

**A composition for
detecting 1st target
nucleic acid
(UnderSC)**

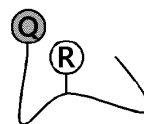
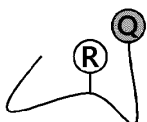
**A composition for
detecting 2nd target
nucleic acid
(OverSC)**

(a) In the absence of a corresponding target nucleic acid

At 1st detection
temperature

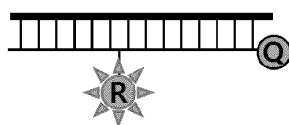


At 2nd detection
temperature

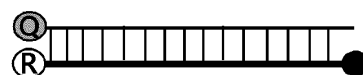


(b) In the presence of a corresponding target nucleic acid

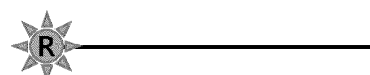
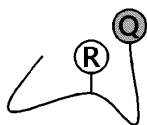
At 1st detection
temperature



Generation of fluorescence



At 2nd detection
temperature



Generation of fluorescence

Ⓡ : Reporter dye

Ⓢ : Quencher

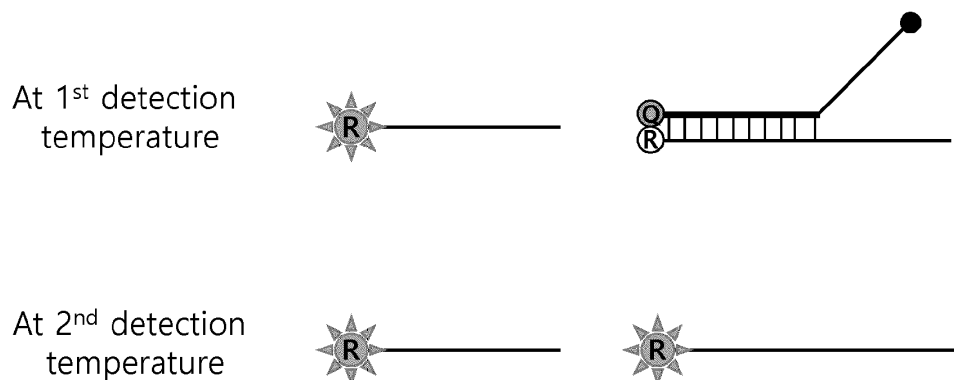
● : Spacer C3

[Fig. 10]

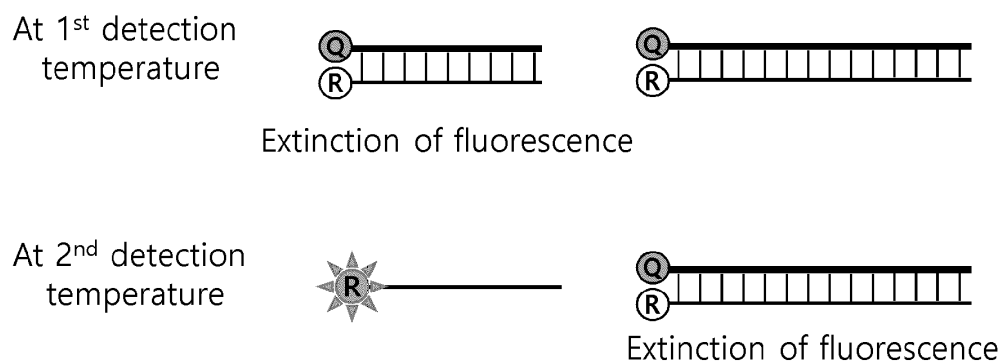
**A composition for
detecting 1st target
nucleic acid
(InterSC)**

**A composition for
detecting 2nd target
nucleic acid
(InterSC)**

(a) In the absence of a corresponding target nucleic acid



(b) In the presence of a corresponding target nucleic acid



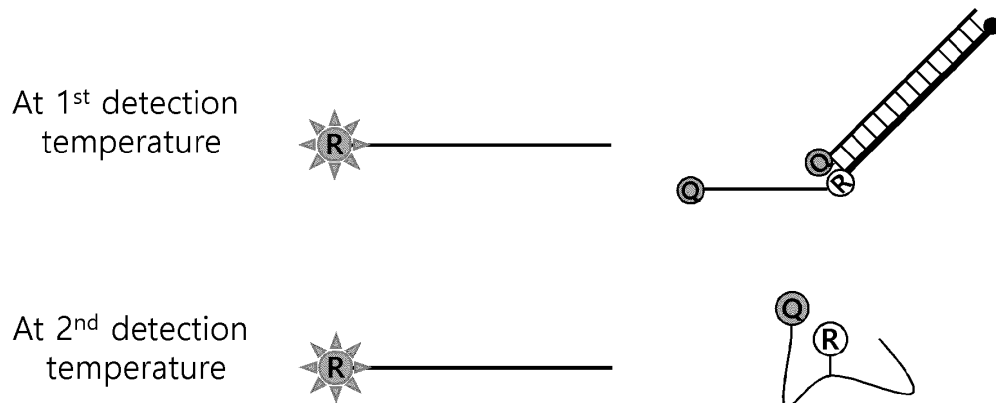
Ⓡ : Reporter dye Ⓢ : Quencher ● : Spacer C3

[Fig. 11]

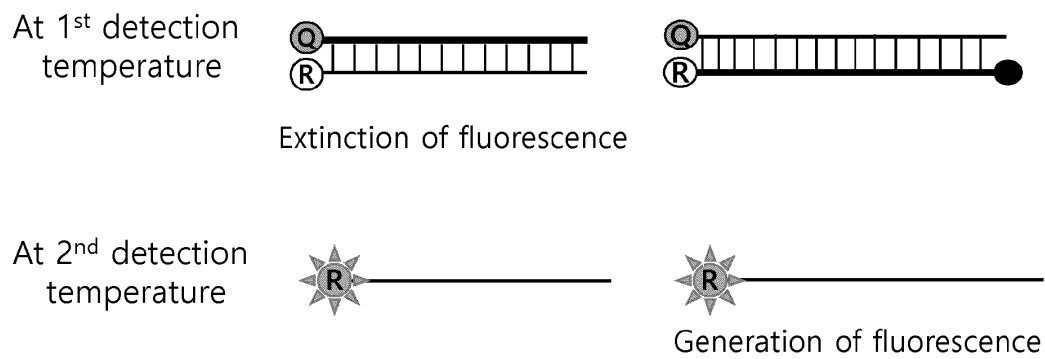
**A composition for
detecting 1st target
nucleic acid
(InterSC)**

**A composition for
detecting 2nd target
nucleic acid
(OverSC)**

(a) In the absence of a corresponding target nucleic acid

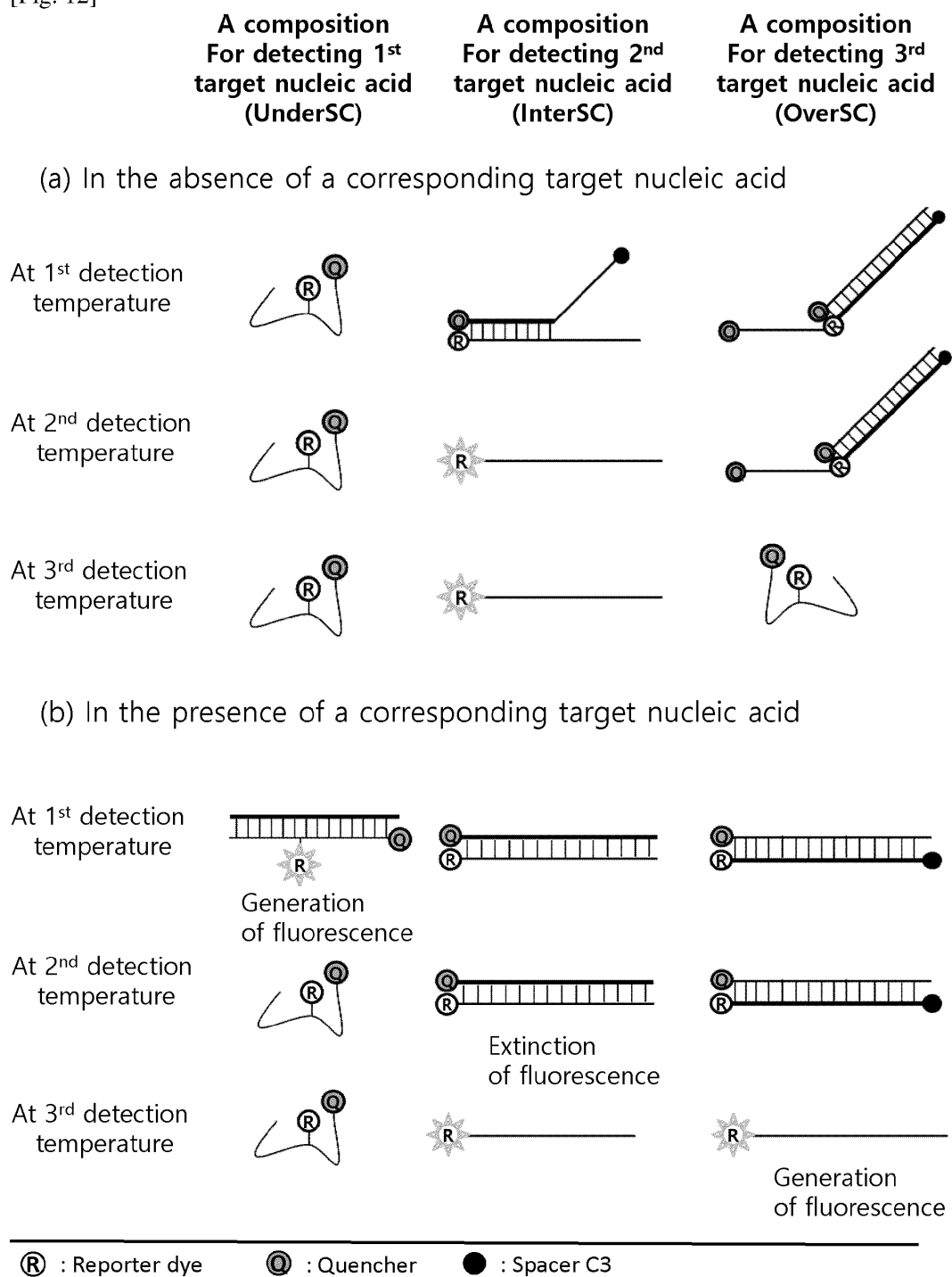


(b) In the presence of a corresponding target nucleic acid

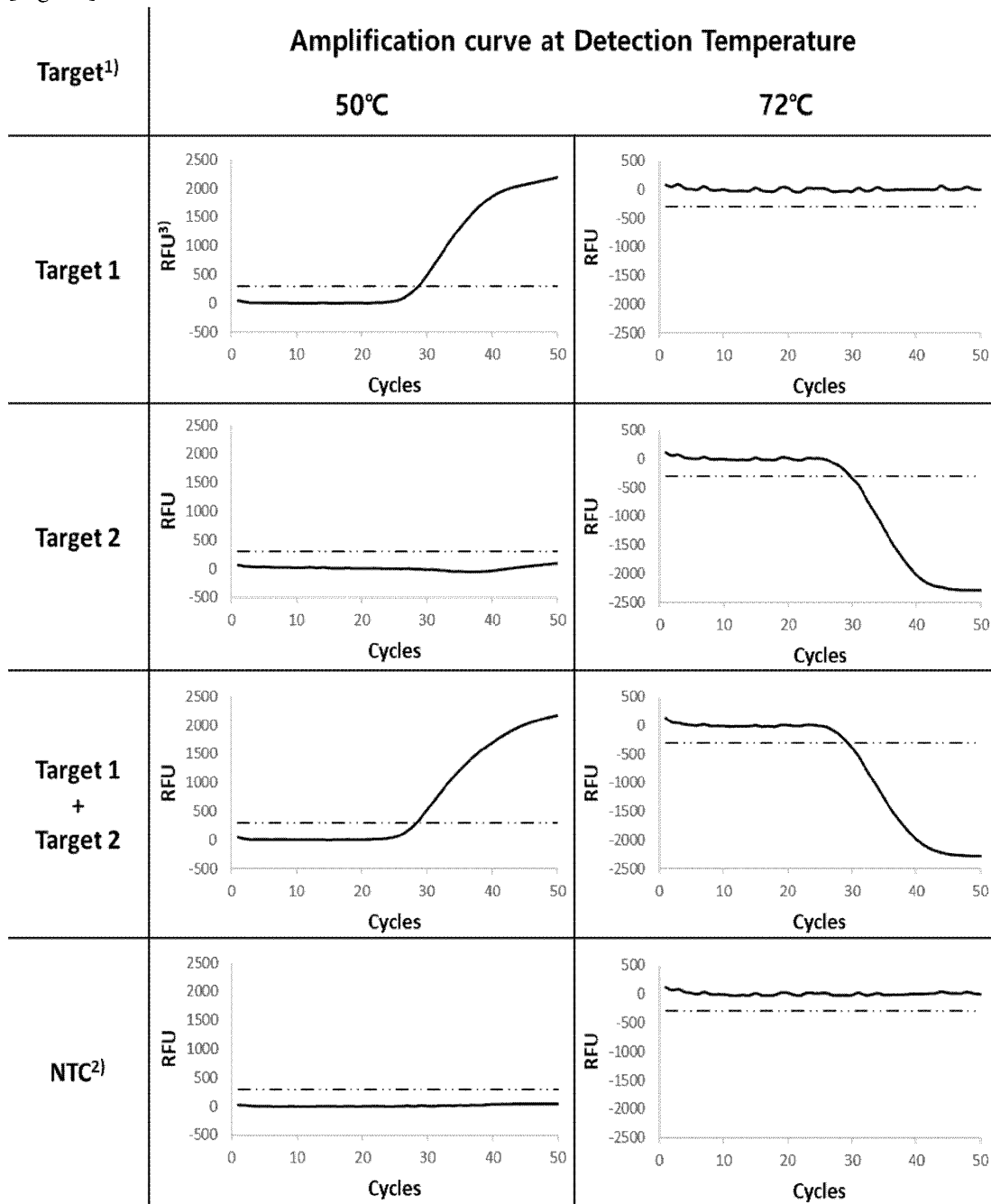


Ⓡ : Reporter dye Ⓢ : Quencher ● : Spacer C3

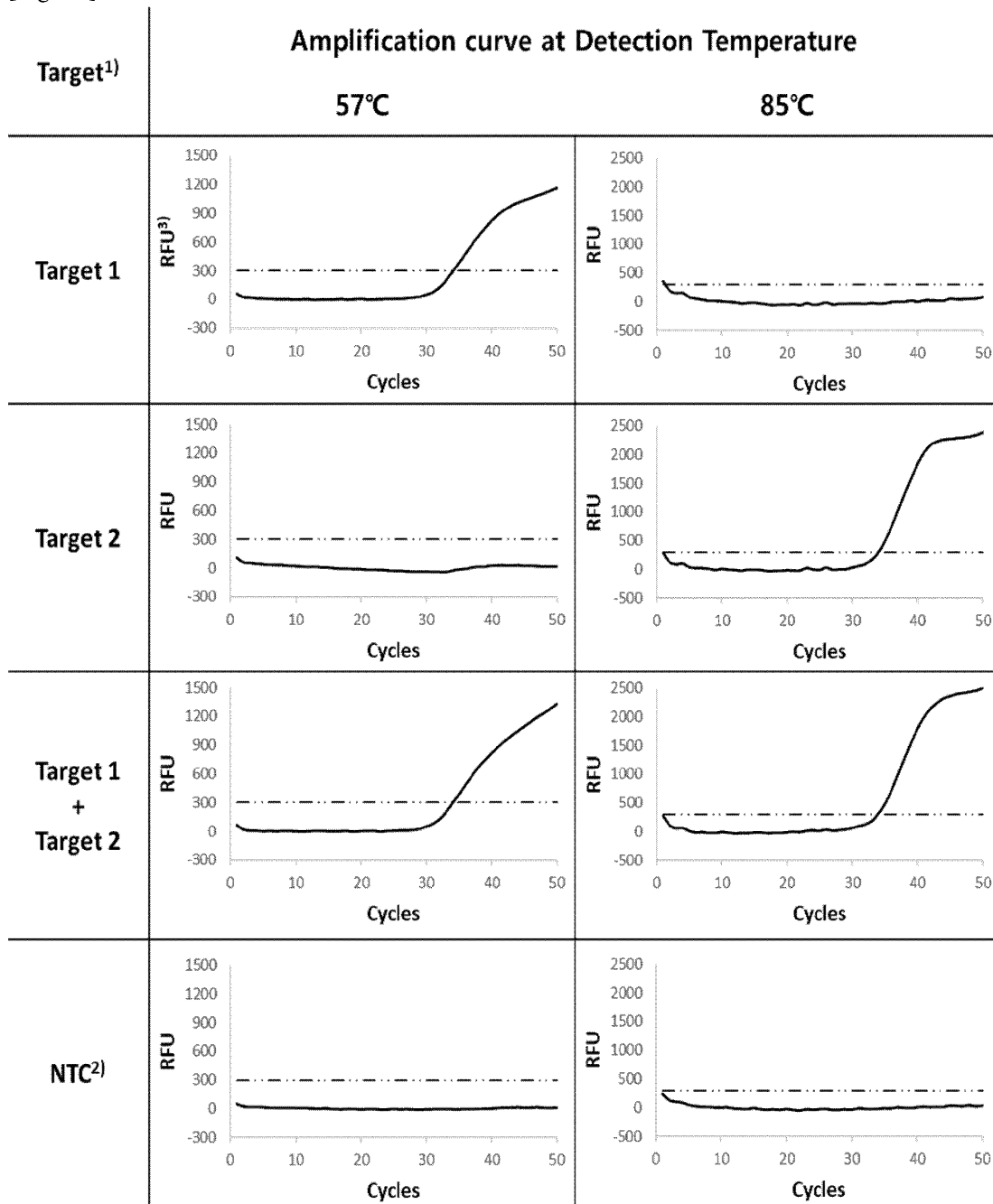
[Fig. 12]



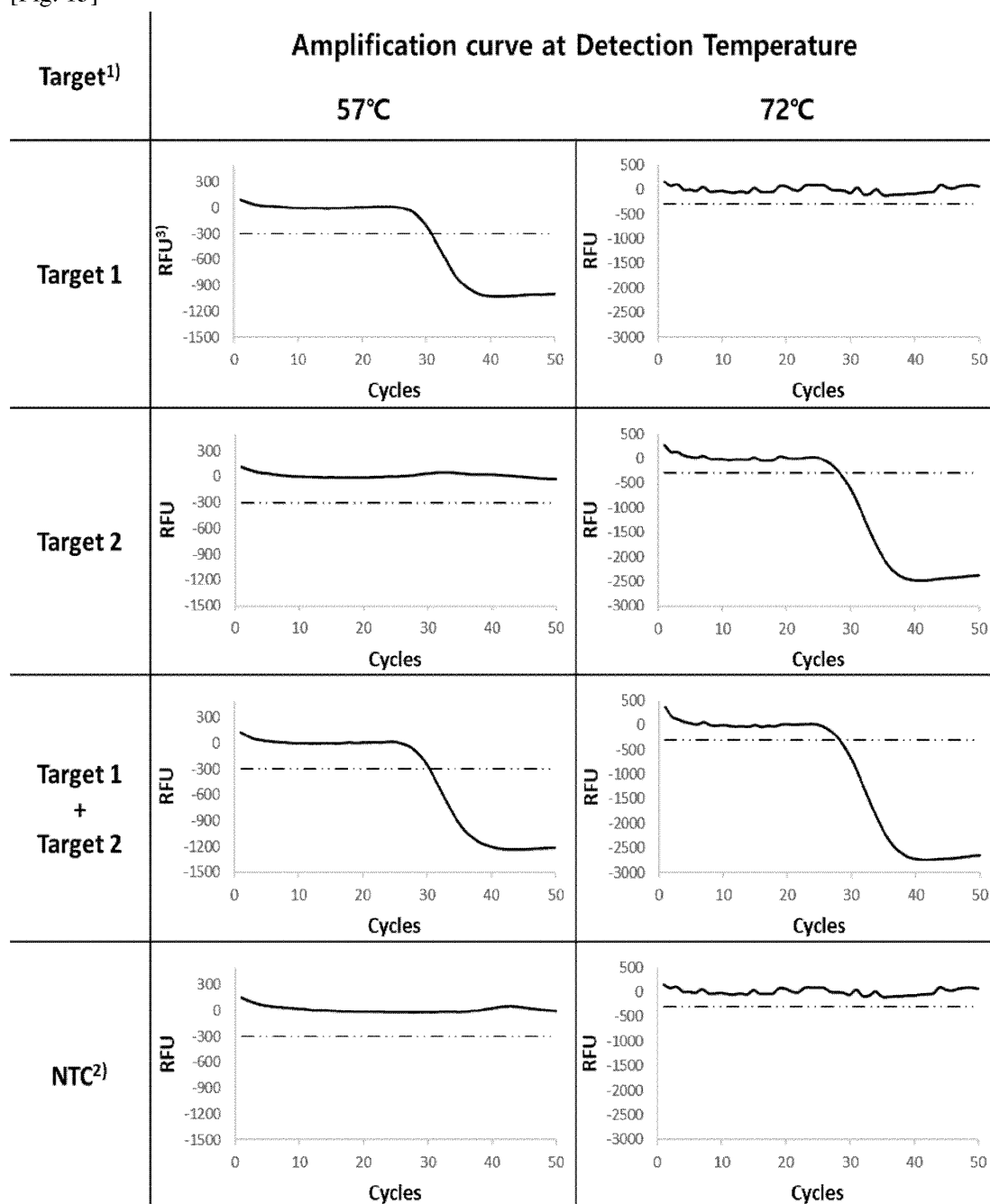
[Fig. 13]



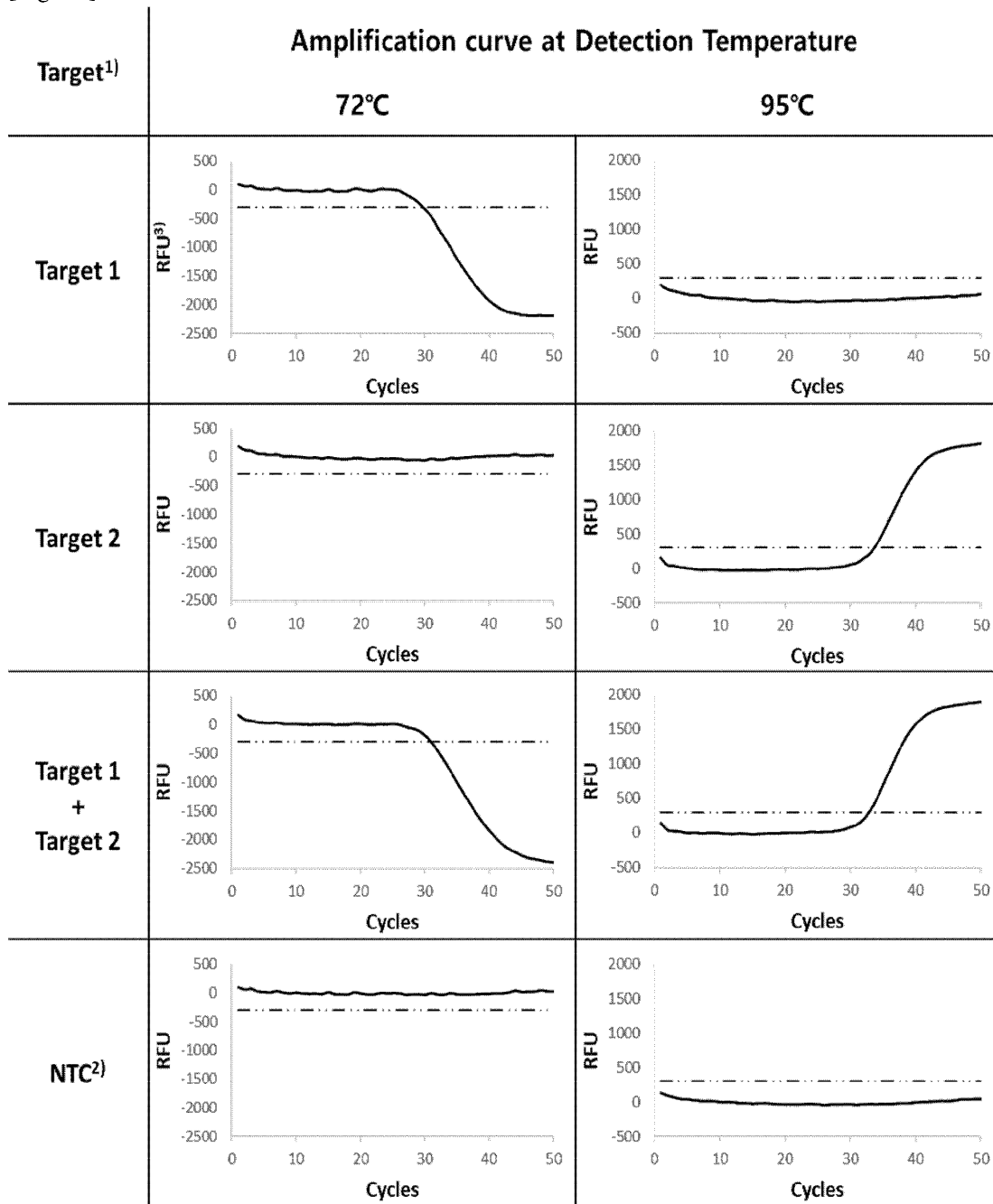
[Fig. 14]



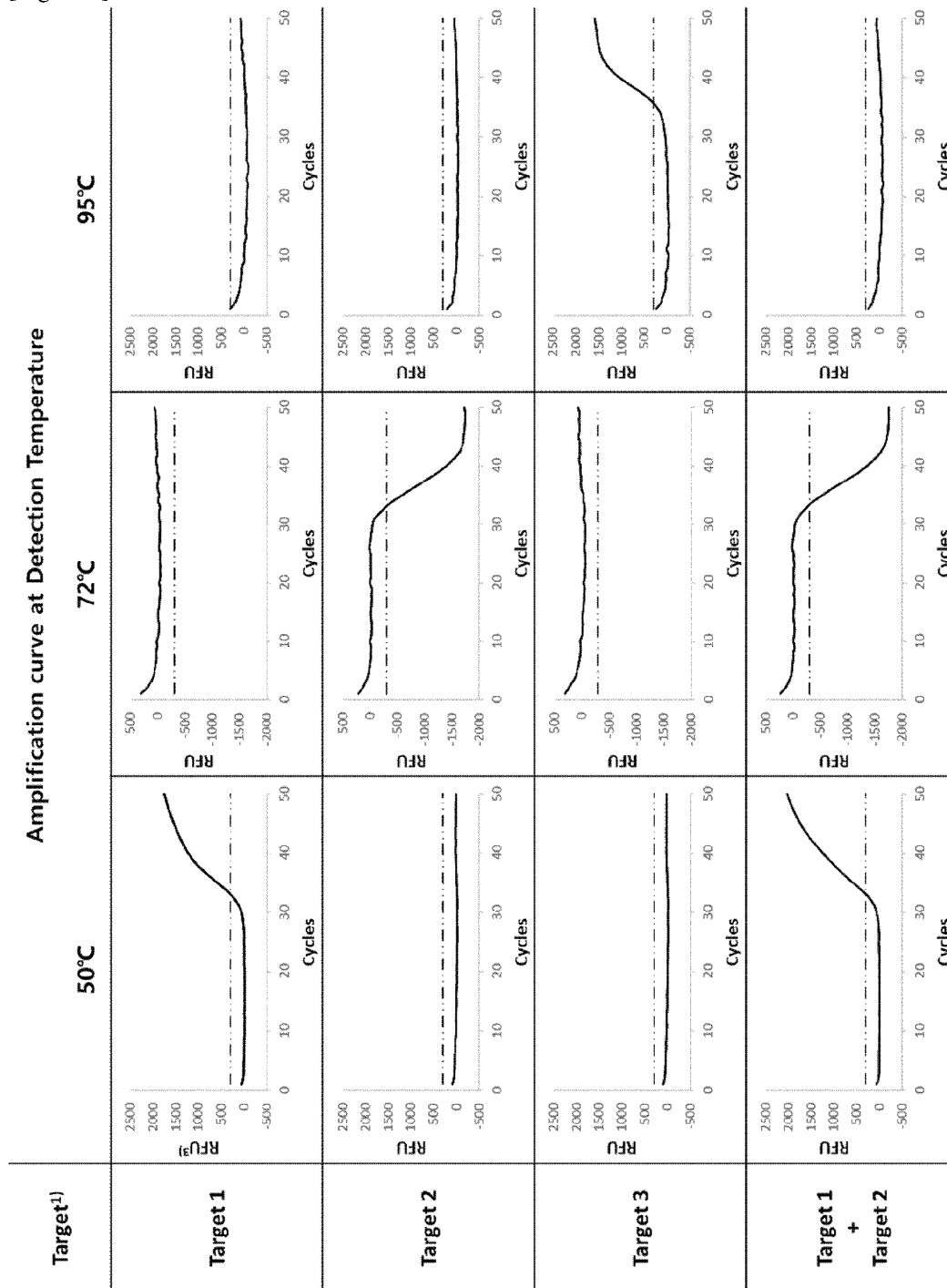
[Fig. 15]



[Fig. 16]



[Fig. 17a]



[Fig. 17b]

