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PCR mixture and condition (2X SUPERGREEN PCR MASTER MIX) V.2

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For generic primers

1 PCR mixture PCR condition for **generic primers**

A	B	C
Component	Volume	Final conc.
Template (< 1 µg)	X µL	-
Forward Primer (10 µM)	1 µL	0.4 µM
Reverse Primer (10 µM)	1 µL	0.4 µM
2x Master Mix	12.5 µL	-
ddH2O	up to 25 µL	-
Total volume	25 µL	-

2 PCR condition for **generic primers**

A	B	C	D
Step	Temp	Sec	Cycle
Initial denaturation	95 °C	180	
Denaturation	95 °C	30	35 cycles
Annealing	53-58 °C	30	
Extension	72 °C	60	
Final extension	72 °C	210	
Preservation	4 °C	∞	

For barcoded primers

3 PCR mixture for **barcoded primers**

A	B	C
Component	Volume	Final conc.
Template (< 1 µg)	X µL	-
Forward Primer (10 µM)	5 µL	2 µM
Reverse Primer (10 µM)	5 µL	2 µM
2x Master Mix	12.5 µL	-
ddH ₂ O	up to 25 µL	-
Total volume	25 µL	-

4 PCR condition for **barcoded primers**

A	B	C	D
Step	Temp	Sec	Cycle
Initial denaturation	95 °C	180	
Denaturation	98 °C	20	35 cycles
Annealing	64 °C		
Extension	72 °C		
Final extension	72 °C	210	
Preservation	Preservation	4 °C	∞