

CURRENT

Category: General SOP to perform Polymerase Chain Reaction (PCR) in Genetics laboratory.
Title: To perform Polymerase Chain Reaction (PCR) in the Department of Clinical Pharmacology, K.E.M Hospital, Mumbai.

SOP No.: PCR Method-02

Date first effective: 1st January 2025

Review date: 31st December 2025

Department of Clinical Pharmacology, 1st floor, New MS building Seth GS Medical College & KEM Hospital, Parel, Mumbai 400012.

Category: General SOP to perform Polymerase Chain Reaction (PCR) in Genetics laboratory.

Title: To perform polymerase chain reaction (PCR) in the Department of Clinical Pharmacology, K.E.M Hospital, Parel, Mumbai.

SOP No.: PCR Method-02

Total pages: 04

Date first effective: 1st January 2025

Review date: 31st December 2025

Version: 03

Author: Dr. Sheetal Kudtarkar
Project Scientist

Signature with date:

Sheetal Kudtarkar
30/12/24

Reviewer1: Dr. Bhaskar Krishnamurthy
Assistant Professor

Signature with date:

BK
30/12/2024

Dr. Bhaskar Krishnamurthy
Assistant Professor,
Department of Clinical Pharmacology,
Seth GSMC and KEMH, Mumbai - 400 012.

Reviewer2: Dr. Mahesh Belhekar
Associate Professor

Signature with date:

Belhekar
30/12/2024

Dr. Mahesh N. Belhekar
Associate Professor
Department of Clinical Pharmacology,
New MS Building, First Floor,
Seth GS Medical College and KEM Hospital,
Acharya Dunde Marg, Parel,
Mumbai - 400 012, India

Approved by: Dr. Nithya Gogtay
Professor and Head

Nithya Gogtay
30/12/24

Signature with date:

Dr. Nithya Gogtay
Professor & Head
Department of Clinical Pharmacology
1st Floor, MS Building,
Seth GS Medical College & KEM Hospital,
Parel, Mumbai - 400 012.

Category: General SOP to perform Polymerase Chain Reaction (PCR) in Genetics laboratory.

Title: To perform Polymerase Chain Reaction (PCR) in the Department of Clinical Pharmacology, K.E.M Hospital, Mumbai.

SOP No.: PCR Method-02

Date first effective: 1st January 2025

Review date: 31st December 2025

Department of Clinical Pharmacology, 1st floor, New MS building Seth GS Medical College & KEM Hospital, Parel, Mumbai 400012.

Table of contents

No.	Contents	Page No.
1	Purpose	3
2	Scope	3
3	Responsibility	3
4	References to other applicable SOPs	3
5	Detailed Instructions	4

Category: General SOP to perform Polymerase Chain Reaction (PCR) in Genetics laboratory.
Title: To perform Polymerase Chain Reaction (PCR) in the Department of Clinical Pharmacology, K.E.M Hospital, Mumbai.

SOP No.: PCR Method-02

Date first effective: 1st January 2025

Review date: 31st December 2025

Department of Clinical Pharmacology, 1st floor, New MS building Seth GS Medical College & KEM Hospital, Parel, Mumbai 400012.

Purpose: This standard operative procedure (SOP) describes the method to perform the Polymerase Chain Reaction (PCR) for amplification of isolated DNA.

Scope: This SOP is limited for performing the Polymerase Chain Reaction.

Responsibilities:

Primary Responsibility: Divya Bhare
Lab Technician

Divya Bhare
30/DEC/2024

Secondary Responsibility: Dr. Sheetal Kudtarkar
Project Scientist

Sheetal Kudtarkar
30/12/24

References: Molecular Cloning
Sambrook & Russell

Category: General SOP to perform Polymerase Chain Reaction (PCR) in Genetics laboratory.

Title: To perform Polymerase Chain Reaction (PCR) in the Department of Clinical Pharmacology, K.E.M Hospital, Mumbai.

SOP No.: PCR Method-02

Date first effective: 1st January 2025

Review date: 31st December 2025

Department of Clinical Pharmacology, 1st floor, New MS building Seth GS Medical College & KEM Hospital, Parel, Mumbai 400012.

Detailed Instructions:

Important : This procedure should be carried out wearing gloves

1. Aliquot 12.5 μL master mix from the stock to setup the PCR reaction for single reaction.
2. To this add 1 μL of each forward and reverse primers in the same tube.
3. Add 3.5 μL of MilliQ water to the same tube and vortex the tube to mix the reaction mixture to make the final volume of 18 μL .
4. Depending upon the no. of samples including negative control and positive control, calculate for the total no. of samples.
5. Now pipette out 18 μL of the above working master mix in different fresh sterile PCR tubes.
6. Keep one tube as negative control i.e. not containing DNA sample and add 2 μL of DNA sample in remaining tube in DNA extraction lab and add known DNA in positive control tube.
7. Add 2 μL of milli Q in negative control tube to make its final volume 20 μL .
8. Now spin the tubes on spinix and keep in Thermal cycler.
9. Set the required temperature condition and carry out the PCR according to **SOP no.PCR**

INS -01.