

SimPCR[™] HPV Screening kit

Cat. No: SBL12-566/50

USER MANUAL

INTRODUCTION

Human Papilloma Virus (HPV) is one of the most common virus groups in the world affecting skin and mucosal areas of human body.

More than 150 HPV types have been identified, 40 of them can infect the genital areas through sexual contact. HPV types are classified according to their association with cancer.

Oncogenic or high risk HPV (HCR) types can cause intraepithelial neoplasia of the anogenital region including cervical, vulvar, vaginal, penile and anal cancers as well as some oropharyngeal cancers. In other hand, low risk HPV types generally cause

genital warts but do not cause cervical cancer.

KIT CONTENTS

Commente	Labala	Volume
Components	Labels	50 Tests
Reaction Mix	Reaction Mix	575 μl
Primer	Oligomix	200 µl
HPV Positive Control	Control Positive	30 µl
HPV Negative Control	Control Negative	30 µl

TEST PRINCIPLE

HPV detection is based on the amplification of the pathogen genome specific region using the HPV specific primers. the product amplification has been confirmed by agarose gel electrophoresis. An internal control primer (IC) in the test tube has confirm the amplification accuracy. The IC has not interfere with the HPV genome amplification.

PROTOCOL

a) Preparation of the PCR mix

For each experiment, prepare a master mix of an appropriate volume for negative control, positive sample, and (n+1) test samples. The reagents have to mix under this ratio:

Component Labels	Volume/reaction	
Reaction Mix	11.5 μl	
Oligomix	4 μΙ	

NOTE: For each PCR set, prepare one positive and one negative controls.

Aliquot 15.5μ l of Master Mix in each tube and $2-5\mu$ l control DNA into individual tubes; then, add DW up to 20 μ l; spin tubes shortly and place them in your thermocycler device.

b) Thermal cycler programming Set device as the following profile:

Step	Temp	Time	Cycle	
Initial activation	95°C	15 min	1 X	
Denaturation	95°C	30 s		
Annealing	53°C	30 s	40 X	
Extension	72°C	30 s		
Final Extension	72°C	5 min	1 X	

Notice: appropriate reaction template files can be found in <u>www.sim-biolab.com</u>

DATA ANALYSIS

Run 10 μ L of PCR products alongside a DNA marker (100bp) on a 2% agarose gel. The HPV render a band of approximately 220bp and internal control primers amplify 500bp fragment.

Band size	Positive result	Positive result	Negative result	Reaction failed (repeat test)
500bp	+	-	+	-
220bp	+	+	-	-

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