

## Bloodborne GPack

(REF: BB/GPACK/100)



## PRINCIPLE OF METHOD

GeneProof PCR kits offer our innovative One Workflow solution allowing the detection of multiple parameters in one run using our Universal PCR Profile for majority of the GeneProof PCR kits (RNA and DNA pathogens).

## WORKFLOW

1. Use the required sample volume in accordance with the extraction manual.
2. Thaw the Universal Internal Control (purple cap) in the refrigerator (2 - 8 °C) for about 15 minutes. Vortex and briefly centrifuge the thawed solution before use. Add Universal Internal Control at 10% of the elution volume to each sample.
3. Perform sample extraction according to the appropriate protocol.
4. Thaw the PCR reagents in a refrigerator (2 - 8 °C) for 15 to 30 min. Vortex and centrifuge briefly before use:

## GeneProof Hepatitis B Virus (HBV) PCR Kit

- HBV Master Mix (blue cap)
- Calibrator A HBV 10<sup>4</sup> IU/μl (black cap)
- Calibrator B HBV 10<sup>3</sup> IU/μl (brown cap)
- Calibrator C HBV 10<sup>2</sup> IU/μl (white cap)
- Calibrator D HBV 10<sup>1</sup> IU/μl (transparent cap)
- Negative control (NFW\* - not included)

## GeneProof Hepatitis C Virus (HCV) Diagnostic PCR Kit

- HCVD Master Mix (blue cap)
- Calibrator A HCVD 10<sup>5</sup> IU/μl (black cap)
- Calibrator B HCVD 10<sup>4</sup> IU/μl (brown cap)
- Calibrator C HCVD 10<sup>3</sup> IU/μl (white cap)
- Calibrator D HCVD 10<sup>2</sup> IU/μl (transparent cap)
- Negative control (NFW\* - not included)

## GeneProof HIV type 1 (HIV-1) Diagnostic PCR Kit

- HIVD Master Mix (blue cap)
- Calibrator A HIVD 10<sup>5</sup> IU/μl (black cap)
- Calibrator B HIVD 10<sup>4</sup> IU/μl (brown cap)
- Calibrator C HIVD 10<sup>3</sup> IU/μl (white cap)
- Calibrator D HIVD 10<sup>2</sup> IU/μl (transparent cap)
- Negative control (NFW - not included)

\*NFW= Nuclease-free water

5. **PCR preparation** (total reaction volume is 40 (HBV) or 50 (HCVD) μl), pipette:

- a) A1: 30 μl HBV Master Mix + 10 μl Calibrator A 10<sup>4</sup> IU/μl
- b) B1: 30 μl HBV Master Mix + 10 μl Calibrator B 10<sup>3</sup> IU/μl
- c) C1: 30 μl HBV Master Mix + 10 μl Calibrator C 10<sup>2</sup> IU/μl
- d) D1: 30 μl HBV Master Mix + 10 μl Calibrator D 10<sup>1</sup> IU/μl
- e) E1: 30 μl HBV Master Mix + 10 μl Negative Control
- f) F1: 30 μl HBV Master Mix + 10 μl extracted DNA/RNA

- a) A1: 30 μl HCVD Master Mix + 20 μl Calibrator A 10<sup>5</sup> IU/μl
- b) B1: 30 μl HCVD Master Mix + 20 μl Calibrator B 10<sup>4</sup> IU/μl
- c) C1: 30 μl HCVD Master Mix + 20 μl Calibrator C 10<sup>3</sup> IU/μl
- d) D1: 30 μl HCVD Master Mix + 20 μl Calibrator D 10<sup>2</sup> IU/μl
- e) E1: 30 μl HCVD Master Mix + 20 μl Negative Control
- f) F1: 30 μl HCVD Master Mix + 20 μl extracted DNA/RNA

## PCR Plate layout

	1	2	3	4	5	6
A	HBV CalA	HCVD CalA	HIVD CalA			
B	HBV CalB	HCVD CalB	HIVD CalB			
C	HBV CalC	HCVD CalC	HIVD CalC			
D	HBV CalD	HCVD CalD	HIVD CalD			
E	HBV NC	HCVD NC	HIVD NC			
F	HBV S1	HCVD S1	HIVD S1			
G	HBV S2	HCVD S2	HIVD S2			
H	...	...	...			

CalA = Calibrator A; CalB = Calibrator B; CalC = Calibrator C;  
CalD = Calibrator D; NC = Negative Control; S1 = unknown sample 1;  
S2 = unknown sample 2

**ATTENTION! Reaction volume must be set to 40 μl.**

- a) A1: 20 μl HIVD Master Mix + 30 μl Calibrator A 10<sup>5</sup> IU/μl
- b) B1: 20 μl HIVD Master Mix + 30 μl Calibrator B 10<sup>4</sup> IU/μl
- c) C1: 20 μl HIVD Master Mix + 30 μl Calibrator C 10<sup>3</sup> IU/μl
- d) D1: 20 μl HIVD Master Mix + 30 μl Calibrator D 10<sup>2</sup> IU/μl
- e) E1: 20 μl HIVD Master Mix + 30 μl Negative Control
- f) F1: 20 μl HIVD Master Mix + 30 μl extracted DNA/RNA

**ATTENTION! Reaction volume must be set to 40 μl.**



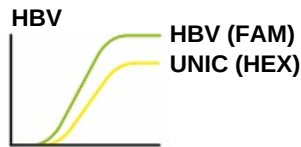
## Bloodborne GPack

- Centrifuge the PCR plate briefly.
- Set up the Universal PCR Profile on the PCR cyclor according to the GeneProof HIV type 1 (HIV-1) Diagnostic PCR Kit.

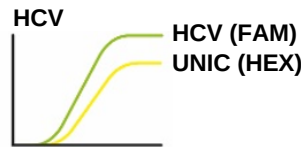
Step	Process	Temperature °C	Time
1	UNG decontamination/ Reverse Transcription	42	15 mins
2	Initial denaturation	95	10 mins
3	Denaturation	95	5 s
	Annealing	60	40 s
	Extension	72	20 s

## EVALUATION

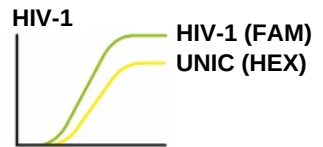
Column 1 - Hepatitis B Virus (HBV)



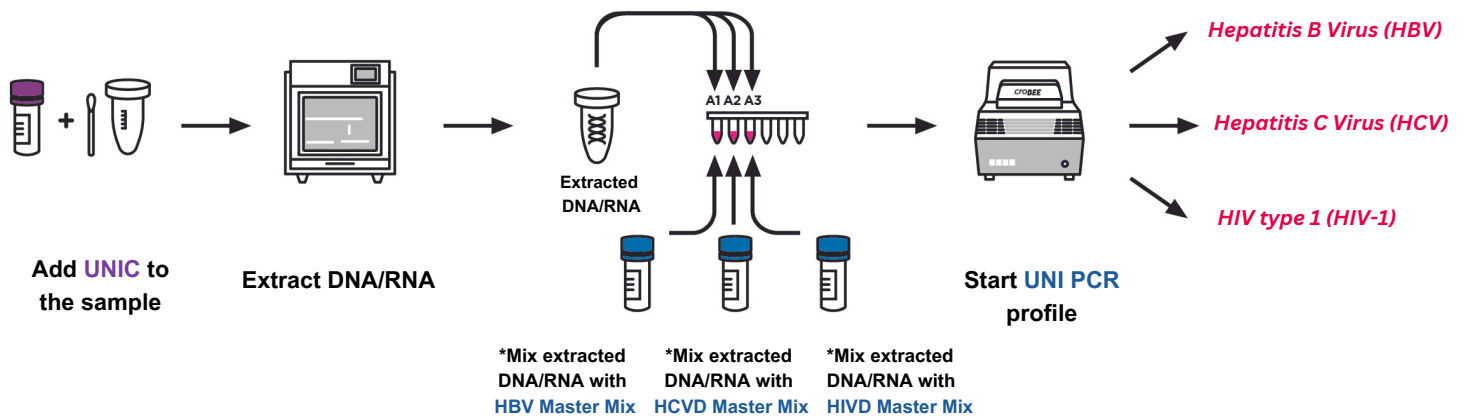
Column 2 - Hepatitis C Virus (HCV)



Column 3 - HIV type 1 (HIV-1)



## Hepatitis GPack Workflow



\*Add appropriate number of Positive and Negative Controls according to the IFU

Choose from the Bloodborne GPacks Portfolio.  
For the results that matter most.

**1. Hepatitis GPack: detection of 2 pathogens**  
Hepatitis B Virus (HBV), Hepatitis C Virus (HCV)

**2. Bloodborne GPack: detection of 3 pathogens**  
Hepatitis B Virus (HBV), Hepatitis C Virus (HCV),  
HIV type 1 (HIV-1)