Epstein-Barr Virus PCR Detection kit

For Research Use Only

Cat. No.: PK3151 **Quantity**: 50 Reactions

Storage:-20°C **Shipment**: Wet/Dry Ice

This kit is destined for qualitative detection of EBV DNA in infected samples by the method of Polymerase Chain Reaction. The reagent of ready to use mix is an optimized 1X PCR mixture of Taq DNA Polymerase (recombinant), PCR buffer, $MgCl_2$, dNTPs and primers. Primer set is specific to the highly specific repetitive region of *BLLF1* gene. This primer set, allows for detection of 30 copies of *Epstein – Barr virus.*

Kit Contents:

1. 1X PCR MIX	1000µl	4. Positive Control	100µl
2. Taq DNA polymerase (5 u/µl)	10µl	5. DNase Free Deionized Sterile Water	3ml
3. Mineral Oil	1ml		

Sample preparation:

DNA Extraction: Performed in Pre-amplification 1, Specimen & Control Area.

DNA can be extracted from Human samples by commercial extraction kits or in-house method.

Human or tissues samples can also be extracted by SinaClon DNA extraction kits: DNP™ (Cat.No.EX6071) or SinaPure™ Viral(Cat.No.EX6061).

PCR Protocol:

PCR Protocol: Performed in Pre- Amplification 2, Reagent Preparation Area.

1. Take out the kit and unfreeze the tubes, then put all the tubes on ice. The final volume of each reaction will be 25µl.
2. Label new PCR tubes for amplification reaction(s) for test(s), positive and negative control.
3. Add the following reagents for each tube on ice:

PCR MIX	19.8 µl / for each reaction
Taq DNA polymerase	0.2 µl / for each reaction

4. To each tube add one drop (20-25 µl) mineral oil (if needed).
5. Add 5 µl DNA*(Use specified pipette for sampling of DNA).
6. Close tubes; spin the mixtures on microfuge for 3-5 sec.
7. Transfer the tubes to preheated thermo cycler and start the program:

Cycling parameters:

Initial Denaturation: 94°C - 3 min

1x then link to

93°C – 30 sec
59°C - 30 sec
72^0C- 30 sec
} 33 Cycles

Final Extension: 72^0C- 5 min 1x

Not: Cycling parameters may need to be setup with some Thermo cyclers.

Resµlt analysis:

Performed in Post –Amplification Area.

In presence of 100bp DNA ladder, analyze 10µl of amplified samples directly in a 2% agarose gel without adding loading buffer. The presence of 239 0r 256bp fragments indicates positive test.

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