

TransDirect® Animal Tissue PCR Kit

Cat. No. AD201

Storage: at -20°C for two years

Description

TransDirect® Animal Tissue PCR Kit uses a unique lysis buffer to lyse animal tissues (fresh or frozen) and blood. The resulting lysate without purification can be directly used as PCR template. 2×TransDirect® PCR SuperMix (+dye) is highly resistant to various PCR inhibitors present in animal tissues. PCR product can be directly used for gel electrophoresis.

Applications

- Direct amplification from unpurified lysate. Suitable for high throughput applications.
- Suitable for mammalian cells, saliva, hair shaft, animal tissues and blood.
- Amplification of genomic DNA fragment up to 3 kb.

Kit Contents

Component	AD201-01	AD201-02
AD1 Buffer	4 ml	20 ml
AD2 Buffer	1 ml	5 ml
AD3 Buffer	4 ml	2×10 ml
2×TransDirect® PCR SuperMix (+dye)	1 ml	5×1 ml
Nuclease-free Water	5 ml	25 ml

Materials

Material	Amount
Mammalian Cells	≤10 ⁶ cell
Hair shaft	≤10 mg
Mouse Tail	≤0.5 cm
Mouse Ear	≤0.5 cm ²
Saliva	≤10 µl
Animal Tissues	≤10 mg
Blood	≤10 µl

Genomic DNA extraction

1. Mix 40 µl of AD1 buffer with 10 µl of AD2 buffer. For more samples, premix AD1 buffer with AD2 buffer at a ratio of 4:1. The mixture can be stored up to 2 hours at room temperature.
2. Sample treatment
 - Mammalian Cells
Pellet the cells by centrifugation and remove the supernatant. Add the mixture of AD1/AD2, mix thoroughly by pipetting up and down.
 - Saliva
Directly add saliva into the mixture of AD1/AD2, mix thoroughly by pipetting up and down.
 - Hair Shafts
Cut hair into pieces, add the mixture of AD1/AD2, mix thoroughly by pipetting up and down.
 - Animal Tissues
Cut up tissues with sterile scissors or blade, add the mixture of AD1/AD2, mix thoroughly by pipetting up and down.
 - Blood
Directly add blood into the mixture of AD1/AD2, mix thoroughly by pipetting up and down.
3. Incubate at room temperature for 10 minutes, followed by at 95°C for 3 minutes (for hard-to-lyse tissues, like hair, we suggest incubating at 55°C for 10 minutes, followed by at 95°C for 3 minutes).

4. Add 40 μ l of AD3 buffer, mix well. The lysate can be used as PCR template or stored at 2-8°C for three months or at -20°C for six months.

Reaction Components

Component	Volume	Final Concentration
Unpurified lysate	Variable ($\leq 4 \mu$ l)	as required
Forward Primer (10 μ M)	0.4 μ l	0.2 μ M
Reverse Primer (10 μ M)	0.4 μ l	0.2 μ M
2 $\times$ TransDirect® PCR SuperMix (+dye)	10 μ l	1 $\times$
Nuclease-free Water	Variable	-
Total volume	20 μ l	-

Thermal cycling conditions

94°C	5-10 min	} 35-40 cycles
94°C	30 sec	
50-60°C	30 sec	
72°C	1-2 kb/min	
72°C	5-10 min	

Notes

- Completely thaw the contents in the tube and mix well before use.
- If faint bands are observed, use more PCR template or increase the number of PCR cycles (no more than 40 cycles).
If non-specific amplification bands are observed, adjust the annealing temperature or properly reduce the quantity of template used.

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