

TransDirect® Animal Tissue PCR Kit

Please read the manual carefully before use.

Cat. No. AD201

Storage: at -20°C for two years

Description

TransDirect[®] Animal Tissue PCR Kit uses a unique lysis buffer to lyse tissues (fresh or frozen). The resulting lysate without purification can be directly used as PCR template. $2 \times TransDirect$ [®] PCR SuperMix (+dye) is highly resistant to various PCR inhibitors present in animal tissues. PCR product can be directly used for gel electrophoresis. AD3 Buffer is recommended to use in aliquots.

Applications

- Direct amplification from unpurified lysate. Suitable for high throughput screening.
- Suitable for mammalian cells, blood, saliva, hair and animal tissues (mammals, marine animals, insects) etc.
- 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

Sample Requirements

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

Procedures

Prepare a 95°C water bath or metal bath in advance.

A. Sample Lysis

1. Mix 40 μ l of AD1 buffer with 10 μ l 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. Different samples, the treatment method is as follows:

a.Mammalian Cells

Completely remove the culture medium from the collected cells. Add the mixture of AD1/AD2, mix thoroughly by pipetting up and down.

b.Animal Tissues

Cut up tissues with sterile scissors or blade, add the mixture of AD1/AD2, mix thoroughly by pipetting up and down.

c.Saliva

Directly add saliva into the mixture of AD1/AD2, mix thoroughly by pipetting up and down.

d.Hair

Cut hair into pieces, add the mixture of AD1/AD2, mix thoroughly by pipetting up and down.





e.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 µl 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.

B. PCR

Reaction Components (20 µl)

Component	Volume	Final Concentration
Unpurified Lysate	Variable (≤4 μl)	as required
Forward Primer (10 µM)	0.4 μl	0.2 μΜ
Reverse Primer (10 μM)	0.4 μl	0.2 μΜ
2×TransDirect® PCR SuperMix (+dye)	10 μl	1×
Nuclease-free Water	Variable	-
Total volume	20 μl	-

PCR

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

Notes

- · Samples avoid repeated freezing and thawing.
- Completely thaw the contents in the tube and mix well before use.
- For tissue that is not easy to lysis (such as hair), it can be incubated at 55°C for 10 minutes.
- 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.
- The extracts can be stored at 2-8°C for three months or at -20°C for six months.