

Instruction for Ten-Minute DNA Release Kit

The DNA kit is designed to obtain DNA extract from mice tail, ear or toe for PCR within 10-30 minutes.


It is a rapid screening method for transgenic mice.

Catalog Number	DNA Release Buffer		Samples	Storage & Use
	Name	Volume		
300-907-004	Buffer A	20 ml	100	-Can be used for 2 years at 20 °C. -or 4 °C for 6 months -Warm to RT before using if it was kept below 4°C.
	Buffer B	4 ml		

Tools

- Thermo-machine or water bath which can be set for a constant temperature between 55° - 95°C;
- Micro Centrifuge;
- 1.5ml micro tubes, pipette tips, and pipette with 10-200ul scale.
-

Protocol

Steps	Action	Example
Step #1	<ul style="list-style-type: none"> • Place 1.5-3.0 mm section of a mouse tail or 2 x3 mm of its ear or toe tissue (or any other tissue** or material stained with body fluid**) in a micro tube. Add 200ul of the Buffer A to the tube. Heat at 93 °C for 3 Min. Cool down 2 Min, Add 40ul Buffer B to the tube. 	
Step #2	<ul style="list-style-type: none"> • Put the tube in a thermomixer machine or water bath at 55° C, shaking at 200-400rpm for 10-30 minutes (If shaking is not available extend the time to 30 minutes*) to release the DNA. * For older mouse tail , you will get a better result in 20-30 minutes 	
Step #3	<ul style="list-style-type: none"> • Flick the tube 3-4 times, heat the tube at 95° C for 5 minutes. 	
Step #4	<ul style="list-style-type: none"> • Centrifuge at 10,000 rpm for 5 min at room temperature. 	
Notes	<p>You may see the part of tail is still in the solution, but the DNA has already been released and it is ready for PCR.</p>	
	<ul style="list-style-type: none"> • Take the 1-2ul DNA extract as the template to run PCR in 20-25 ul reaction volume. • Transfer the rest of DNA extract to a fresh tube at 4 °C. The DNA can be used for 2-4 weeks, or to be kept at -20 °C for long time storage. 	
	<ul style="list-style-type: none"> • Usually, the kit works well for 100-1200 bp PCR fragment. • If your PCR fragment is over 1200 bp, or if you want to get high yield of the DNA to do Southern et al, you should use the Ten-Minute DNA Release Kit-2 to obtain the purified DNA within 1 hour. The purified DNA can be used in both the Southern and PCR. Please visit www.crystalgen.com for details. 	

****Using Different Organs' Tissue or Materials Stained by Body Fluid**

1. Cut 1-3mm³ (1-12 mg) of the sample;
2. If the sample is clean (ex. clear skin tissue), follow the protocol;
3. If the sample is stained with blood or any other dirty trace:
 - a. rinse it 2-3 times with PBS without Ca⁺⁺ and Mg⁺⁺;
 - b. dry the tissue on a paper towel; then, to follow the work table.

Ordering information

Name	Cat. #	Price/Kit	Reactions
Ten-Minute DNA Extraction Kit	300-907-004	\$ 129/Kit	100
		\$ 1100.0 /10 Kits	10 x 100

Reference:

1. Lee TY et al.; Phylogenetic analysis by RFLP and sequencing of mitochondrial DNA in a Korean Population. Arch Oharm Res.,2006 Jan; 29(1) :88-95.
2. Chakraborty A et al.; Molecular identification of hairtail species(Pisces:Trichiuridae) base on PCR-RFLP analysis the mitochondrial 16S rRNA gene. J. Appl. Genet. 2005;46(4): 381-385.
3. Ren S et al. ;A Simplified Method to Prepare PCR Template DNA for Screening of Transgenic and knockout Mice. Contemp Top Lab Anim Sci.2001: 40(2): 27-30.
4. Hofstetter JR et al.; A Comparison of Recovery Methods and Tissue Sources. Biochem Mol Med. 1997 ; 62(2); 197-202.
5. Malumbres M. et al. Isolation of High Molecular Weight DNA for Reliable Genotyping of Transgenic Mice. Bio Techniques 1997; 22(6): 1114-1119.
6. Anderson S. et al.; Sequence and organization of human mitochondrial genome, Nature, 1981; 290(9):457-465.