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PCRBIO Rapid Extract PCR Kit

Product description:

PCRBIO Rapid Extract PCR Kit combines rapid DNA extraction with fast, highly specific DNA amplification in a convenient, easy to use format. Eliminate the need for laborious and time-consuming DNA extraction methods with this simple, integrated extraction and amplification PCR kit powered by the latest advances in hot-start polymerase technology.

PCRBIO Rapid Extract PCR Kit has been developed for fast, efficient amplification of DNA from a variety of tissues and is particularly suited to solid tissue such as mouse tail or mouse ear. DNA extraction is performed in a single tube, removing the need for multiple washing steps. Extraction of DNA is rapid, providing DNA for PCR in 15 minutes. Extraction takes place in a single tube, minimizing potential contamination.

Extracted DNA is amplified using PCRBIO HS Taq Mix Red. Our antibody hot-start polymerase uses the latest developments in polymerase technology and buffer chemistry to enhance PCR speed, yield and sensitivity. The completed reaction is ready for direct gel loading without the need to add loading buffer.

Component	80 reactions	400 reactions
5x PCRBIO Rapid Extract Buffer A	1 x 1.6ml	5 x 1.6ml
10x PCRBIO Rapid Extract Buffer B	1 x 800µl	5 x 800µl
2x PCRBIO HS Taq Mix Red	2 x 1.0ml	10 x 1.0ml

Shipping and storage

On arrival the kit should be stored between -30˚C and -15˚C. If stored correctly the kit will retain full activity for 12 months. The kit can go through 30 freeze/thaw cycles with no loss of activity.

Limitations of product use

The product may be used only for in vitro research purposes.

Technical support

For technical support and troubleshooting please email technical@pcrbio.com the following information:

Reaction setup

Screen grabs of qPCR or PCR data.

Sample amounts

Sample	Amount per 100µl extraction	Notes
Mouse tail clip	1 to 2mm (2.5 to 6mg)	
Mouse ear punch	2 to 4mm² (2.5 to 6mg)	
Animal tissue	3 to 30mg	
Hair follicle	1-10 individual follicles	
Buccal swab	1 swab	Use 300ul extraction volume for higher yield
Mammalian blood	2 to 8ul Fresh/EDTA blood	2mm² FTA, FTA elute or Guthrie cards
FFPE tissue	1mm³ or 2mm² of 10µm section	

Protocol

1. Extraction reaction setup

For each biological sample, create the following 100µl extraction reaction:

Reagent	100µl reaction	Notes
Mouse tail clip	1 to 2mm (2.5 to 6mg)	See table above for other samples
5x PCRBIO Rapid Extract Buffer A (1u/µl)	20µl	Lysis buffer
10x PCRBIO Rapid Extract Buffer B	10µl	Protease containing buffer
PCR grade dH_2O	70µl	

2. Extraction reaction incubation

Incubate extraction reaction for lysis, nuclease and protein denaturation, followed by heat-inactivation:

Cycles	Temperature	Time	Notes
1	75˚C	5min	Vortex twice during incubation
1	95˚C	10min	Deactivates protease

3. Dilute then centrifuge reaction

Add 900µl PCR grade dH_2O to the deactivated reaction. Centrifuge at high speed in a microcentrifuge for 1 minute to pellet debris. Supernatant can be used directly in PCR or stored between -30˚C and -15˚C.

4. PCR Reaction setup

Prepare a master mix based on the following table:

Reagent	50µl reaction	Final concentration
2x PCRBIO HS Taq Mix Red	25.0µl	1x
Forward primer (10µM)	2.0µl	400nM
Reverse primer (10µM)	2.0µl	400nM
Supernatant from step 3	1.0µl to 2.0µl	variable
PCR grade dH_2O	Up to 50µl final volume	

Cycle using conditions based on the following table:

Cycles	Temperature	Time	Notes
1	95˚C	1min to 2min	Initial denaturation and enzyme activation. For colony PCR increase denaturation time to 10 minutes
40	95˚C 55˚C to 65˚C 72˚C	15 seconds 15 seconds 1 to 90 seconds	Denaturation Anneal Extension (15 seconds per kb). For multiplex PCR use 90 seconds

Analyse by agarose gel electrophoresis, the reaction contains a red dye for tracking during electrophoresis. In a 2% agarose TAE gel the dye migrates at a rate equivalent to 350bp of DNA. In a 1% agarose TAE gel the dye migration rate is equivalent to 600bp of DNA.