

DNA/RNA Extraction Kit

CATALOGUE NUMBER	KIT SIZE (TESTS)
PCREXT1	50T

The DNA/RNA Extraction Kit comprises reagents which together are used to isolate and purify DNA or RNA from biological specimens and cultured cells. The resulting purified DNA or RNA is suitable for subsequent applications such as real time quantitative PCR.

Test Principle:

Biological samples or cultured cells are lysed by addition of Lysing Buffer to release DNA and RNA. Magnet beads are also added to the sample tube and the released DNA and RNA binds to these beads. The lysate is placed next to a magnet which holds the nucleic acid coated beads against the side of the tube while the Lysing Buffer and impurities are removed by washing. The purified DNA and RNA is eluted in buffer.

Reagents:

The kit comprises five bottles of reagents of the following composition:

REAGENT	SIZE
Lysing Buffer	1 x 20 ml
Wash Buffer 1	1 x 30 ml
Wash Buffer 2	1 x 30 ml
Eluting Buffer	1 x 5 ml
Magnetic Beads	1 x 1.3 ml

Materials not provided: Heating block (65°C, adjustable), Class II biological safety cabinet, vortex, microfuge, nuclease-free centrifuge tubes, micropipettes and sterile, nuclease free pipette tips

Precautions:

For in vitro diagnostic use by trained professionals only.

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Follow Good Laboratory Practice procedures where samples and kits are handled and treat the kit components and all samples as if potentially infectious. Follow local regulations for correct disposal of samples, unused reagents, used kit components and assay waste.

Wear protective clothing including laboratory coat, disposable gloves and safety glasses

when conducting the test. Change gloves often to prevent contamination of samples and reagents.

Do not use any component of the kit after the expiry date. Do not incorporate reagents from different kit batches or reagents from other commercially available kits.

The Lysing Buffer contains a guanidine salt which can form highly reactive products when combined with sodium hypochlorite. Do not add bleach or acidic solutions to reaction tubes

or liquid washing waste. Suitable detergent may be added.

Avoid any of the reagents coming into contact with skin, eyes and mucous membranes. If accidental contact should occur, flush immediately with a copious amount of water.

Do not reuse plastic laboratory consumables.

Storage and Stability:

The kit should be stored in dry conditions at room temperature (15 – 30° C). The kit is stable up to the expiry date printed on the outer label.

Sample Collection and Preparation:

The DNA/RNA Extraction Kit can be used on respiratory samples, swabs, saliva and nasopharyngeal or oropharyngeal secretion samples stabilised in storage solution. Ideally DNA or RNA extraction from the samples should be performed immediately. Alternatively, the samples may be stored up to 24 hours at $2-8^{\circ}\text{C}$ or for longer at -70°C. Avoid repeat freeze-thawing.

Assav Procedure:

Sample Pre-treatment

- For respiratory samples and saliva, vortex the samples to mix them then pipette 200 μ l into a new 1.5 ml nuclease-free centrifuge tube. For swab samples, cut the tip of the swab into a 1.5 ml nuclease-free centrifuge
- tube and add 1 ml of Normal Saline (0.9%). Vortex to mix and extract the sample then pipette 200 μ l into a new centrifuge tube.
- For samples stabilised in storage reagent, vortex the samples to mix then pipette 200 μ l into a new 1.5 ml nuclease-free centrifuge tube.

Before beginning the extraction procedure make sure the reagent bottles are intact and without any leaks. Shake the reagent bottles gently to mix the contents.

- Add 400 μl Lysing Buffer and 25 μl Magnetic Beads to each sample tube, close the lids, vortex the tubes for 10 seconds to mix and then briefly centrifuge the tubes for 5 - 10 seconds.

- Place the tubes in a dry heating block and incubate at 60°C for 15 minutes.

 Mix the contents of the tubes by repeated vortexing.

 Place tubes in a magnetic separation apparatus to magnetise the Mag-Bind particles until the supernatant becomes clear (approximately 3 minutes).
- Discard the supernatants from the tubes then remove the tubes from the magnetic separation apparatus. Add $600\,\mu$ I Wash Buffer 1 to each tube, close the lids and vortex to mix for 10
- Place tubes in the magnetic separation apparatus until the buffer becomes clear (approximately 3 minutes). Discard the supernatant buffer from the tubes then take out the tubes from the magnetic separation device. Important: Remove as much of the waste buffer as possible.
- Add 600 μl Wash Buffer 2 to each tube, close the lids and vortex to mix for 10
- Place tubes in the magnetic separation apparatus until the buffer becomes clear (approximately 3 minutes). Discard the supernatant buffer from the tubes then take out the tubes from the magnetic separation device. **Important:** Remove as much of the waste buffer as possible.

- Add 100 µl Eluting Buffer to each tube, close the lids and mix by vortexing for 10 seconds. Put the tubes in a heating block and incubate at 65°C for 5 minutes. Mix the contents of the tubes by vortexing briefly.
- Place the tubes in the magnetic separation apparatus until the eluent becomes clear (approximately 3 minutes). Transfer the eluent to a fresh nuclease-free
- The purified DNA/RNA in the eluent can be used immediately for up to 2 hours or store at -70°C.

Limitations of the Kit:

The DNA/RNA Extraction Kit is only to be used by laboratory personnel trained in PCR

The quality of the DNA or RNA is affected by factors including source of sample, sample

collection process, collection site and storage conditions.

Sample quality has a significant impact on the quality and yield of purified DNA or RNA. The presence of DNA/RNase in the laboratory environment may cause the degradation of the DNA and RNA during or after the purification process. All equipment, consumables and the workbench should be treated before starting to ensure all surfaces are DNA/RNase free.

Symbols:

REF	Catalogue number	\mathcal{A}	Temperature limitation
(i	Consult instructions for use	LOT	Batch code
IVD	In vitro diagnostic medical device	¥	Use by Date
	Manufacturer	(2)	Do not reuse
200	Keep away from sunlight		



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AMS UK Ltd 42 Ballymena Business Centre, Galgorm, Co. Antrim, BT42 1FL, United Kingdom. Tel: +44 (0) 28 25656268