

Apoptotic DNA Ladder Kit

Cat. No. 11 835 246 001 20 tests

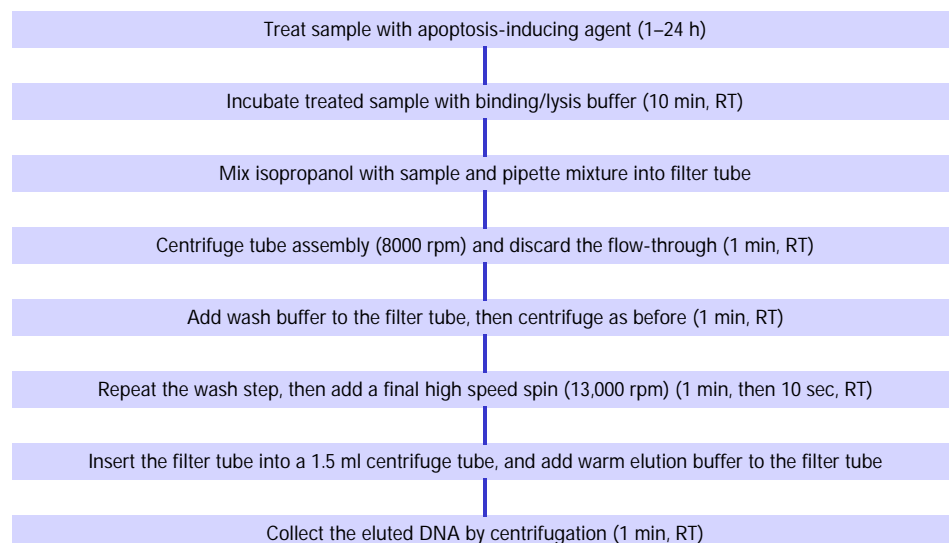
Type	DNA purification kit
Useful for	Preparation of apoptotic DNA fragments for display on electrophoretic gels
Samples	Whole blood or cells in culture
Method	Cell lysis, followed by binding of cellular DNA on glass fiber, removal of impurities, and DNA recovery
Time	DNA preparation: < 20 min (after induction of apoptosis)

Significance of kit: This kit offers the easiest way to isolate apoptotic DNA fragments for DNA ladder analysis. The purification method outlined in the kit is much faster than other DNA purification methods (*e.g.*, phenol/chloroform extraction, DNA precipitation). Purified DNA may be mixed directly with gel loading buffer and analyzed on an agarose gel.

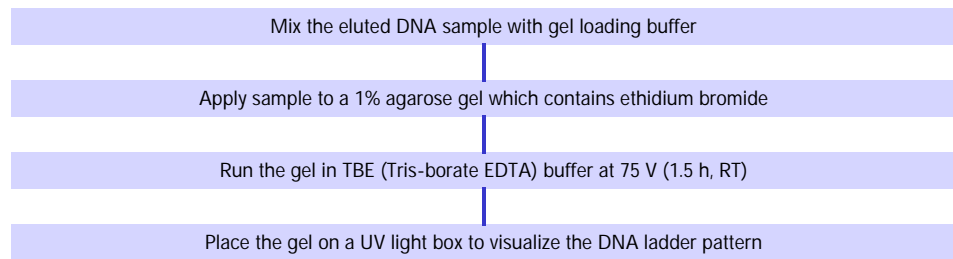
Test principle: Apoptotic DNA binds quickly to glass fiber fleece in the presence of a chaotropic salt, guanidine hydrochloride (guanidine HCl). After cellular impurities are washed off the fleece, the DNA is released from the fleece with a low salt buffer. The procedure (see Flow Chart 1) involves:

- 1 Incubating an aliquot of apoptotic cells with an equal volume of binding/lysis buffer. After the incubation, the lysed sample is poured into a filter tube containing glass fiber fleece.
- 2 Using centrifugation to separate the DNA in the lysate (which binds to the glass fiber fleece) from unbound lysate components (which flow through the fleece into a collection tube).
- 3 Washing the bound DNA twice.
- 4 Eluting the purified DNA from the filter tube and collecting it by centrifugation.

Sample Preparation



DNA Ladder Assay



▲ **Flow Chart 1: Assay procedure, Apoptotic DNA Ladder Kit.**

Sample size: 200 – 300 µl whole blood or cell suspension (for instance, 2 x 10⁶ cells). The kit allows simultaneous processing of multiple samples.

Yield

Sample	Sample volume	Yield of purified DNA
Whole blood (human)	200 µl	3–6 µg
Cultured cells (K562)	2 x 10 ⁶ cells	10 µg

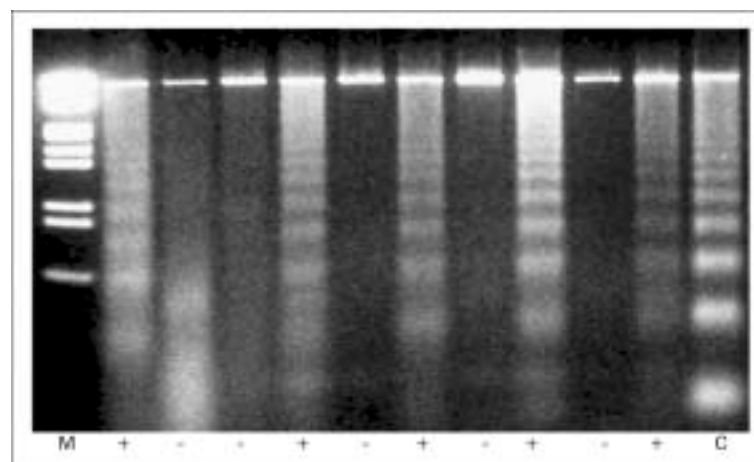
Specificity: Only nucleic acid will bind to the glass fiber filters under the conditions outlined in the kit. Salts, proteins, and other cellular components do not bind.

Kit contents

1. Nucleic acid binding/lysis buffer, ready-to-use
2. Washing buffer (ethanol to be added before use)
3. Elution buffer, ready-to-use
4. Glass fiber filter tubes, 700 µl capacity
5. Polypropylene collection tubes, 2 ml (for washes)
6. Positive control, apoptotic U937 cells, lyophilized

Typical results: see Figure 6

Other applications: For more examples of how the Apoptotic DNA Ladder Kit can be used in the lab, see Appendix, pages 128–129.



▲ **Figure 6: DNA ladder assayed with the Apoptotic DNA Ladder Kit**

- M = Size marker
- = Control cells without camptothecin
- + = Cells treated with camptothecin
- C = Positive control from the kit