Isolation of DNA from Gels

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The technique of isolating DNA from a gel has many applications including the isolation of individual DNA fragments to be used for subcloning a specific piece of DNA. DNA isolated from bands in gels can also be digested with additional restriction endonucleases to generate restriction endonuclease maps. Individual bands of DNA can also be labelled and used as specific probes in hybridization experiments.

As gel electrophoresis is used more often in the undergraduate laboratory, there is a need for experiments demonstrating further applications of gel electrophoresis. Gels can be used in Southern blotting experiments (Karcher, 1991). Gels can also be used to isolate specific DNA fragments. This mini workshop demonstrated how to isolate DNA from agarose gels by electroelution.

To isolate specific fragments of DNA from an agarose gel by gel electroelution, the DNA is first digested with the appropriate restriction endonuclease and subjected to electrophoresis through an 0.8% agarose gel. After electrophoresis, the gel is stained in ethidium bromide (0.5 μg/ml) and viewed under ultraviolet light to visualize the DNA bands. Using great care to minimize exposure to the ultraviolet light, the specific DNA band of interest is cut out of the gel with a razor blade or scalpel. Care is taken not to scratch the surface of the ultraviolet light box. Placing the gel on a sheet of plastic wrap on top of the light box can minimize scratches to the light box. Using forceps and gloved hands, the gel slice with the DNA of interest is placed inside a dialysis tubing bag. The bag is filled with a small amount of gel electrophoresis running buffer (0.5X TBE; 0.0445 M Tris—Trizma base—pH 8.0, 0.0445 M boric acid, 0.01 M EDTA—ethylenediamine tetraacetic acid) and tied or clamped shut. The dialysis bag is then placed in an electrophoresis chamber and subjected to electrophoresis until the DNA migrates out of the gel piece. The DNA migrates out of the agarose gel fragment and remains in the buffer within the dialysis tubing. The dialysis bag is then opened and the buffer (which contains the DNA) within the dialysis bag is removed with a pipet. The buffer-DNA sample can be used as is or the sample can be precipitated with alcohol to concentrate the DNA. The DNA sample can be further purified by phenol extraction. The DNA isolated this way can be used in additional restriction endonuclease digestion reactions or in labelling reactions. For more details, see Karcher (1991:12).

Electroelution is a useful method to isolate DNA from gels in the teaching laboratory because it is an easy procedure that allows the students to visualize the DNA readily during the procedure. There are numerous other methods to isolate DNA from gels, including centrifugation through filter paper. For some examples, see the references listed below.

Ericson, M. L. 1990. Quick DNA recovery from agarose gels by ultracentrifuge run. Trends in Genetics, 6:278.

Heery, D. M., F. Gannon, and R. Powell. 1990. A simple method for subcloning DNA fragments from gel slices. Trends in Genetics, 6:173. (Centrifugation through glass wool.)

Karcher, S. J. 1991. Non-radioactive DNA hybridization experiments for the undergraduate laboratory: The Southern blot analysis. Pages 1–31, *in* Tested studies for laboratory teaching. Volume 12. (C. A. Goldman, Editor). Proceedings of the 12th Workshop/Conference of the Association for Biology Laboratory Education (ABLE), 218 pages.

Koenen, M. 1989. Recovery of DNA from agarose gels using liquid nitrogen. Trends in Genetics, 5:137.

Peloquin, J. J., and E. G. Platzer. 1991. A simple inexpensive electroelution device for the recovery of nucleic acid fragments from agarose gels. BioTechniques, 10:159–160. (Electroelution using a microfuge tube instead of dialysis bag.)

Vogelstein, B., and D. Gillespie. 1979. Preparative and analytical purification of DNA from agarose. Proceedings of the National Academy of Sciences, U.S.A., 76:615–619. (Use of chaiotropic salt NaI.)

Weichenhan, D. 1991. Fast recovery of DNA from agarose gels by centrifugation through blotting paper. Trends in Genetics, 7:109.