

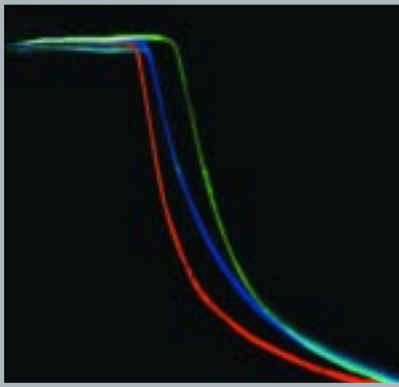
A Mutation Detection Technique Using the FMBIO®

Denaturing Gradient Gel Electrophoresis

DGGE is an extremely sensitive technique capable of discriminating among sequence variants that differ by as little as a single nucleotide. To do this, small PCR products (200-700 bp) are run through an urea/

formamide gradient acrylamide gel. As the fragments enter higher denaturing environments, they begin to denature. Depending on their sequence, mobility shifts are observed as the partially denatured fragments migrate. Gels are then analyzed as though the differences in allele positions are based on size.

Multiple samples on a perpendicular DGGE



Data provided by Dr. K. Miller at the Pacific Biological Station.

Combine The Power of Fluorescence with DGGE

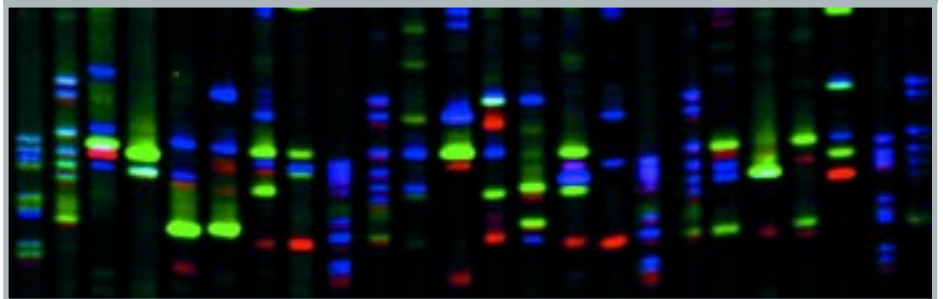
Using the FMBIO and the new FM-DGGE technique, complex loci with multiple melting domains can be analyzed on a single gel with repeated scanning. Fluorescence also eliminates the tedious gel-staining procedure, which saves resources and increases throughput. Perpendicular DGGE determinations become a snap by combining the labeled DNA from multiple samples.



Increase Laboratory Productivity

The FMBIO can significantly increase your laboratory's throughput and reduce sample processing costs by multiplexing DGGE reactions. Multicolor detection and separation capabilities of the FMBIO mean that up to four colors can be utilized in each lane. Less sample can be loaded in each lane due to the superior sensitivity of fluorescence, offering further savings. PCR volumes can now easily be scaled down to 10 µl. Additionally, since the gel plates are never separated, thinner and faster gels can be run without the concern of gel breakage.

Simultaneous detection of 48 samples with FM-DGGE on the FMBIO



Coho salmon (Oncorhynchus kisutch) from three populations were analyzed using 6-Fam, Hex and Rox at a MHC locus. Collaborating with Dr. K. Miller, we have developed a novel DGGE system that will allow researchers to run 52 lanes per gel. Using this technique increases sample throughput to 156 samples per gel.

Advantages

- Dramatically increase sample throughput by multiplexing
- Scan two gels in four colors in under 15 minutes
- FMBIO software offers rapid and accurate analysis of multiple gels with user-defined parameters
- Using fluorescence PCR volumes can be scaled down to 10 μ l
- No staining means no gel breakage
- Run thinner gels faster
- Multiple melting domains can be analyzed on a single gel
- The FMBIO scanning format is compatible with any DGGE system

Reference:

Miller, K., Ming, T., Schulze, A., Bucklin, K. and Calavetta, M.J. 2000. Denaturing Gradient Gel Electrophoresis (DGGE): A Rapid and Sensitive Technique to Screen Nucleotide Sequence Variation in Populations. In: *Polymorphism Analysis and Detections Techniques*, Eds. Burczak, J. and Mardis, E. Eaton Publishing. 550 pp.

FMBIO II Specifications

Laser:	Solid-state 532 nm YAG laser
Scan Area and Time:	20 cm x 43 cm dual color at standard resolution: 10 minutes
Detection Wavelengths:	Up to 4-color separation, selectable from 500 nm to 700 nm
Dynamic Range:	4 orders of magnitude
Compatible Dyes:	For a complete list please visit our Web site
Multiplex Dye Sets (filter):	<ul style="list-style-type: none">• 6-Fam (505 nm), Hex (585 nm) , Texas Red (650 nm)• 6-Fam (505 nm), Hex (560 nm), Ned (585 nm), Rox (605 nm)• Filters for novel dye sets are also available

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