A regularly spaced DNA size standard with 10 kbp resolution for pulsed field gel electrophoresis

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In pulsed field gel electrophoresis (PFGE) a lambda ladder (~50 kbp multiples) is a common DNA size standard (1). Here we describe a standard to conveniently size DNA between ladder rungs and to control for anomalies in PFGE DNA mobility. We combine two partial digests of lambda ladders to give a regularly spaced standard with ~10 kbp resolution in the 10 to 200 kbp range.

METHOD Lambda ladder in solution is partially digested with *Apa* I (~0.1 x unit activity) or *Nae* I (~1.0 x unit activity). These digests are stored at 4° C with 20 mM EDTA pH 7.5 and 1% SDS. Enzymes and lambda DNA are from New England Biolabs.

The figure shows lambda ladder and BRL high molecular weight standard (~1 µg total DNA) in the left lane and, in the right lane, an equal mixture of *Apa* I and *Nae* I partial digests of lambda ladder (~2 µg total DNA). Fragment maps and sizes (kbp) are shown to the right (see ref. 2 for lambda maps). In each lambda unit (48.5 kbp), the left vertical line marks the *Apa* I site and the other vertical line marks the *Nae* I site. Bands with the sizes of intact lambda ladder are also produced by two restriction cuts within one lambda polymer (maps not shown). Bands corresponding to intact lambda ladder are more intense and provide convenient reference points. Electrophoresis is in 1.0% agarose, 0.5 x TBE at 200 V and 14° C with 2 to 15 s switch times over 24 hours on a Bio-Rad CHEF-DR system.

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REFERENCES 1. Carle, G. F. and Olson, M. V. (1984) Nucl. Acids Res. 12, 5647-5664. 2. New England Biolabs catalog 1988-1989.