Promega Notes Magazine **Number 47, 1994, p.15**

Restriction Enzyme Information Resources

Mary Hoenecke, Carolyn Stock
Promega Corporation

Numerous complementary resources provide the researcher with detailed information regarding restriction enzymes, their physical properties and applications in molecular biology. This information is generally presented in the form of concise reviews, tables and charts, making it easy for a scientist to find the necessary data. Additional sources which are of use with restriction enzymes include a number of databases and sequence analysis software packages as well as vendor materials. This article summarizes some of these standard resources.

Restriction endonuclease reviews

Several excellent review articles that convey a wealth of information concerning the properties of restriction enzymes are available. These references cover the commercial availability of restriction enzymes, their general properties such as sequence recognition, star activity, methylation sensitivity, and basic procedures for use of restriction enzymes. Most of the reviewers follow the restriction enzyme nomenclature established by Smith and Nathans (1). Two of these reviews stand out above the others: Roberts and Macelis (2) and Kessler and Manta (3).

Roberts and Macelis

Roberts and Macelis annually publish "REBASE-restriction enzymes and methylases" in *Nucleic Acids Research* (2), which is likely the most referenced source of restriction enzyme information. This compilation is also available as a database and can be obtained at no extra charge through electronic mail (see below). The information presented in the Roberts and Macelis review is updated daily, and these updates are incorporated monthly. Specialized versions are available.

This important reference contains an enzyme listing detailing the recognition sequence and the microorganism from which each enzyme is isolated. Additional information includes commercial sources of enzymes, any known isoschizomers or neoisoschizomers, and methylation sensitivities of the enzymes, if known. A separate table presents recently identified restriction enzymes and provides a published reference. For previously identified restriction enzymes, a published reference source may be found in earlier editions of this review.

Kessler and Manta

An alternate assembly of restriction enzyme endonuclease information has been compiled by Kessler and Manta and appears in *Gene* (3). The Kessler and Manta reference contains information particularly useful for developing cloning strategies, presented in extensive tables. Tables listing isoschizomers, compatible restriction enzyme overhangs, and generation of new sites through enzymatic treatments of restriction enzyme overhangs represent some of the material covered. While the reference is not updated as regularly as the Roberts review, an appendix reports new restriction endonucleases and methylases yearly.

Other restriction enzyme reviews also exist. Bhagwhat (4), Brooks (5), Fuchs and Blakesley (6), Wells *et al*. (7), and Szybalski *et al*. (8) may be of particular interest.

Methylation reviews

While the Roberts publication is considered the definitive reference for restriction enzymes, Nelson *et al*. have published an exhaustive summary covering methylases (9). This review also appears in *Nucleic Acids Research* and is updated every few years. This publication covers methylation sensitivity of restriction endonucleases, DNA methyltransferases and their specificities, and the methylation sensitivity of Type II DNA methyltransferases. In addition, isoschizomer/isomethylator pairs that differ in their sensitivity to sequence-specific methylation are indicated.

DNA methylases are also reviewed in the Kessler and Manta reference discussed above. For more information on troubleshooting problems related to methylation, please see reference 10.

Computer databases and computer software

An increasing number of databases and software have become available and provide information on restriction enzymes, DNA sequences and restriction mapping. In addition, newer software programs identify sequences that may be changed to create new restriction sites without breaking constraints such as amino acid sequence or a consensus protein-binding site. For a variety of organisms, theoretical predictions of site frequency based on known base composition and repetitive sequences are being compiled. This will be useful for selecting restriction endonucleases for experiments requiring megabase-sized fragments.

REBASE

REBASE is the definitive restriction enzyme database containing a listing of all known restriction enzymes, their associated methylases and isoschizomers. Relevant abstracts from the literature are also included. REBASE is available in numerous formats including an ASCII text file arranged in fields for easy formatting. Many of these files are designed specifically for use with specific software programs such as GCG, IGSuite, GENEPRO and Staden. On-line access to this database is available through the GenBank(TM), NCBI and EMBL File Servers, and monthly updates are available through e-mail or by anonymous ftp from vent.neb.com (192.138.220.2). REBASE is maintained by R. J. Roberts and D. Macelis, and a summary of it is published by them yearly in *Nucleic Acids Research* (see above).

For more information on REBASE, contact:

Dr. Richard J. Roberts
New England Biolabs, Inc.
32 Tozer Road
Beverly, MA 01915
e-mail: roberts@neb.com

GenBank and EMBL data libraries

The GenBank Sequence Data Bank, sponsored by the National Institutes of Health and managed by the National Center for Biotechnology Information (NCBI), and the EMBL Data Library, sponsored by the European Molecular Biology Laboratory, are computerized depositories of all known DNA sequences. Sequence data submitted to either data bank is exchanged with the other regularly. Reference 11

explains the process of submitting sequences to these data banks.

For more information, contact:

NCBI
Bldg. 38A, Room 8N-803
8600 Rockville Pike
Bethesda, MD 20894
e-mail: info@ncbi.nlm.nih.gov

EMBL Data Library
European Molecular Biology Laboratory
Postfach 10.2209
Meyerhofstrasse 1
D-6900 Heidelberg, Germany
e-mail: datalib@embl-heidelberg.de

Sequence analysis software

Numerous computer software packages can provide useful alternatives to traditional restriction enzyme analyses. These various packages contain a variety of programs, including simple restriction enzyme site identification, creation of linear and circular restriction maps, and mutagenesis strategies to create new restriction sites. For more details regarding the programs within a particular software package, please contact the vendor. Some vendors include Genetics Computer Group, Intelligenetics®, DNASTAR®, Hitachi and International Biotechnologies.

While each of these packages provides detailed instructions for use of the software, two additional references covering computerized sequence analysis may be useful. These are *Sequence Analysis Primer* (11) and *Sequence Analysis in Molecular Biology: Treasure Trove or Trivial Pursuit* (12).

Vendor materials

Many suppliers of molecular biological products produce catalogs and other resources which contain useful data covering restriction enzyme characteristics. The Promega 1993/1994 Catalog, for example, contains tabular information on restriction enzyme isoschizomers, restriction enzyme overhang compatibility, heat susceptibility and reaction buffer compositions, making usage of our restriction enzymes more convenient. Similar tables can also be found in Promega's *Protocols and Applications Guide*. In addition, Promega's Technical Services Scientists are available to troubleshoot any problems encountered when using restriction enzymes in a particular experiment.

Summary

A variety of resources provide scientists with helpful information regarding the use of restriction enzymes. Familiarity with these sources offers researchers the ability to effectively plan experiments and to solve potential problems.

References

1. Smith, H.O. and Nathans, D. (1973) *J. Mol. Biol.* **81**, 419.
2. Roberts, R.J. and Macelis, D. (1993) *Nucl. Acids Res.* **21**, 3125.

3. Kessler, C. and Manta, V. (1990) *Gene* **92**, 1.
4. Bhagwat, A.S. (1992) *Meth Enzymol.* **216**, 199.
5. Brooks, J.E. (1987) *Meth. Enzymol.* **152**, 113.
6. Fuchs, R. and Blakesley, R. (1983) *Meth. Enzymol*. **100**, 3.
7. Wells, R.D., Klein, R.D. and Singleton, C.K. (1981) In: *The Enymes* **14,** Boyer, P.D., ed, Academic Press, San Diego, 158.
8. Szybalski, W. *et al*. (1991) *Gene* **100**, 13.
9. Nelson, R.M., Raschke, E. and McClelland, M. (1993) *Nucl. Acids Res.* **21**, 3139.
10. Schoenfeld, T. (1993) *Promega Notes* **42**, 22.
11. Gribskov, M. and Devereux, J. (1991) *Sequence Analysis Primer*, Stockton Press, NY.
12. von Heijne, G. (1987) *Sequence Analysis in Molecular Biology: Treasure Trove or Trivial Pursuit.* Academic Press, San Diego.

For more information on Promega's Restriction Enzymes, please request the 1994 Restriction Enzyme Product Guide.
