

**Patent Number:** 

[11]

#### US005858671A

## United States Patent [19]

[54] ITERATIVE AND REGENERATIVE DNA

#### Jones

5,858,671

**Date of Patent:** Jan. 12, 1999 [45]

[]	SEQUENCING METHOD		
[75]	Inventor:	Douglas H. Jones, Iowa City, Iowa	
[73]	Assignee:	The University of Iowa Research Foundation, Iowa City, Iowa	
[21]	Appl. No.:	742,755	

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[22]	Filed:	Nov. 1, 1996
[51]	Int. Cl. <sup>6</sup>	

**U.S. Cl.** ...... **435/6**; 435/91.1; 435/91.2 

#### [56] References Cited

#### U.S. PATENT DOCUMENTS

5,102,785	4/1992	Livak et al	435/6
5,403,708	4/1995	Brennan et al	435/6
5,508,169	4/1996	Deugau et al	435/6
5,552,278	9/1996	Brenner	435/6
5,599,675	2/1997	Brenner	435/6
5,604,097	2/1997	Brenner	435/6
5,695,934	12/1997	Brenner	435/6
5,714,330	2/1998	Brenner et al	435/6
5,763,175	6/1998	Brenner	435/6

#### FOREIGN PATENT DOCUMENTS

WO 91/02796	3/1991	WIPO .
WO 92/15711	9/1992	WIPO .
WO 93/21340	10/1993	WIPO .
WO 95/27080	10/1995	WIPO .
WO 96/12039	4/1996	WIPO .

#### OTHER PUBLICATIONS

Straus et al., Proc. Natl. Acad. Sci. USA 87, 1889-1893 (1990).

Jones, BioTechniques 22, 938-946 (1997).

Baxter, G. et al., "Microfabrication in Silicon Microphysiometry," Clin. Chem., vol. 40, No. 9, 1800-1804 (1994). Beattie, K. et al., "Advances in Genosensor Research," Clin. Chem., vol. 41, No. 5, 700-706 (1995).

Brenner, S. and Livak, K., "DNA Fingerprinting by Sampled Sequencing," Proc. Natl. Acad. Sci. USA, vol. 86, 8902-8906 (1989).

Broude, N. et al., "Enhanced DNA Sequencing by Hybridization," Proc. Natl. Acad. Sci. USA, vol. 91, 3072-3076

Burns, M. et al., "Microfabricated Structures for Integrated DNA Analysis," *Proc. Natl. Acad. Sci. USA*, vol. 93, 5556-5561 (1996).

Caetano-Anollés, G. et al., "Primer-Template Interactions During DNA Amplification Fingerprinting with Single Arbitrary Oligonucleotides," Mol. Gen. Genet., vol. 235, 157-165 (1992).

Canard, B. and Sarfati, R.S., "DNA Polymerase Fluorescent Substrates with Reversible 3'-tags," Gene, vol. 148, 1-6 (1994).

Carrano, A.V. et al., "A High-Resolution, Fluorescence--Based, Semiautomated Method for DNA Fingerprinting," Genomics, vol. 4, 129-136 (1989).

Cheng, J. et al., "Chip PCR. II. Investigation of Different PCR Amplification Systems in Microfabricated Silicon--Glass Chips," Nucleic Acids Research, vol. 24, No. 2, 380-385 (1996).

Chetverin, A. and Kramer, F., "Oligonucleotide Arrays: New Concepts and Possibilities," BioTechnology, vol. 12, 1093-1099 (1994).

Davis, L. et al., "Rapid DNA Sequencing Based Upon Single Molecule Detection," Genetic Analysis, Techniques, and Applications, vol. 8, No. 1, 1-7 (1991).

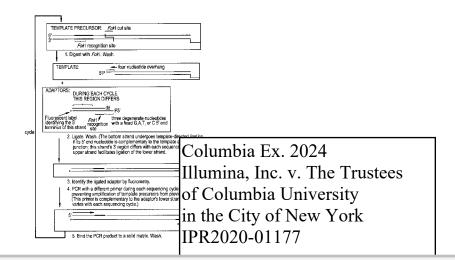
Drmanac, R. et al., "DNA Sequence Determination by Hybridization: A Strategy for Effecient Large-Scale Sequencing," Science, vol. 260, 1649–1652 (1993).

Eggers, M. and Ehrlich, D., "A Review of Microfabricated Devices for Gene-Based Diagnostics," Hematologic Pathology, vol. 9, No. 1, 1–15 (1995).

Eggers, M. et al., "A Microchip for Quantitative Detection of Molecules Utilizing Luminescent and Radioisotope Reporter Groups," BioTechniques, vol. 17, No. 3, 516-524 (1994).

Gibbs, R. et al., "Identification of Mutations Leading to the Lesch-Nyhan Syndrome Automated Direct DNA Sequencing of In Vitro Amplified cDNA," Proc. Natl. Acad. Sci. USA, vol. 86, 1919-1923 (1989).

Green, E. and Green, P., "Sequence-tagged Site (STS) Content Mapping of Human Chromosomes: Theoretical Considerations and Early Experiences," PCR Methods and Applications, vol. 1, 77-90 (1991).







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[73]	Assignee:	The University of Iowa Research Foundation, Iowa City, Iowa
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[56]		References Cited

#### U.S. PATENT DOCUMENTS

5,102,785 5,403,708 5,508,169	4/1995 4/1996	Livak et al	435/6 435/6
	4/1996 9/1996 2/1997 2/1997		435/6 435/6 435/6 435/6
5,714,330 5,763,175	2/1998	Brenner et al	435/6

#### FOREIGN PATENT DOCUMENTS

WO 91/02796	3/1991	WIPO .
WO 92/15711	9/1992	WIPO .
WO 93/21340	10/1993	WIPO .
WO 95/27080	10/1995	WIPO .
WO 96/12039	4/1996	WIPO .

#### OTHER PUBLICATIONS

Straus et al., Proc. Natl. Acad. Sci. USA 87, 1889-1893 (1990).

Jones, BioTechniques 22, 938-946 (1997).

Baxter, G. et al., "Microfabrication in Silicon Microphysiometry," Clin. Chem., vol. 40, No. 9, 1800–1804 (1994). Beattie, K. et al., "Advances in Genosensor Research," Clin. Chem., vol. 41, No. 5, 700-706 (1995).

Brenner, S. and Livak, K., "DNA Fingerprinting by Sampled Sequencing," *Proc. Natl. Acad. Sci. USA*, vol. 86, 8902–8906 (1989). Broude, N. et al., "Enhanced DNA Sequencing by Hybridization," Proc. Natl. Acad. Sci. USA, vol. 91, 3072-3076

Burns, M. et al., "Microfabricated Structures for Integrated DNA Analysis," *Proc. Natl. Acad. Sci. USA*, vol. 93, 5556-5561 (1996).

Caetano-Anollés, G. et al., "Primer-Template Interactions During DNA Amplification Fingerprinting with Single Arbitrary Oligonucleotides," Mol. Gen. Genet., vol. 235, 157-165 (1992).

Canard, B. and Sarfati, R.S., "DNA Polymerase Fluorescent Substrates with Reversible 3'-tags," Gene, vol. 148, 1-6 (1994).

Carrano, A.V. et al., "A High-Resolution, Fluorescence--Based, Semiautomated Method for DNA Fingerprinting," Genomics, vol. 4, 129-136 (1989).

Cheng, J. et al., "Chip PCR. II. Investigation of Different PCR Amplification Systems in Microfabricated Silicon--Glass Chips," Nucleic Acids Research, vol. 24, No. 2, 380-385 (1996).

Chetverin, A. and Kramer, F., "Oligonucleotide Arrays: New Concepts and Possibilities," BioTechnology, vol. 12, 1093-1099 (1994).

Davis, L. et al., "Rapid DNA Sequencing Based Upon Single Molecule Detection," Genetic Analysis, Techniques, and Applications, vol. 8, No. 1, 1-7 (1991).

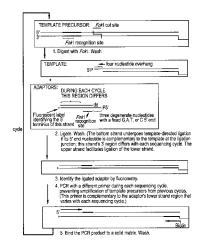
Drmanac, R. et al., "DNA Sequence Determination by Hybridization: A Strategy for Effecient Large-Scale Sequencing," Science, vol. 260, 1649–1652 (1993).

Eggers, M. and Ehrlich, D., "A Review of Microfabricated Devices for Gene-Based Diagnostics," Hematologic Pathology, vol. 9, No. 1, 1–15 (1995).

Eggers, M. et al., "A Microchip for Quantitative Detection of Molecules Utilizing Luminescent and Radioisotope Reporter Groups," BioTechniques, vol. 17, No. 3, 516-524 (1994).

Gibbs, R. et al., "Identification of Mutations Leading to the Lesch-Nyhan Syndrome Automated Direct DNA Sequencing of In Vitro Amplified cDNA," Proc. Natl. Acad. Sci. USA, vol. 86, 1919–1923 (1989).

Green, E. and Green, P., "Sequence-tagged Site (STS) Content Mapping of Human Chromosomes: Theoretical Considerations and Early Experiences," PCR Methods and Applications, vol. 1, 77-90 (1991).





Gyllensten, U. and Allen, M., "PCR-based HLA Class II Typing," *PCR Methods and Applications*, vol. 1, 91–98 (1991).

Gyllensten, U. and Erlich, H., "Generation of Single–Stranded DNA by the Polymerase Chain Reaction and its Application to Direct Sequencing of the HLA–DQA Locus," *Proc. Natl. Acad. Sci. USA*, vol. 85, 7652–7656 (1988).

Han, J. and Rutter, W., "\lambdagter graphs a Phage Expression Vector for the Directional Cloning of cDNA by the use of a Single Restriction Enzyme Sfil," *Nucleic Acids Research*, vol. 16, No. 24, 11837 (1988).

Jacobs, K. et al., "The Thermal Stability of Oligonucleotide Duplexes is Sequence Independent in Tetraalkylammonium Salt Solutions: Application to Identifying Recombinant DNA Clones," *Nucleic Acids Res.*, vol. 16, 4637–4650 (1988).

Kikuchi, Y. et al., "Optically Accessible Microchannels Formed in a Single–Crystal Silicon Substrate for Studies of Blood Rheology," *Microvascular Research*, vol. 44, 226–240, (1992).

Kobayashi, M. et al., "Fluorescence-based DNA Minisequence Analysis for Detection of Known Single-base Changes in Genomic DNA," *Molecular and Cellular Probes*, vol. 9, 175–182 (1995).

Kohsaka, H. and Carson, D., "Solid-Phase Polymerase Chain Reaction," *Journal of Clinical Laboratory Analysis*, vol. 8, 452–455 (1994).

Kricka, L. et al., "Imaging of the Chemiluminescent Reactions in Mesoscale Silicon–Glass Microstructures," *J Biolumin Chemilumin*, vol. 9, 135–138 (1994).

Kuppuswamy, M. et al., "Angle Nucleotide Primer Extension to Detect Genetic Diseases: Experimental Application to Hemophilia B (Factor IX) and Cystic Fibrosis Genes," *Proc. Natl. Acad. Sci. USA*, vol. 88, 1143–1147 (1991).

Lagerkvist, A. et al., "Manifold Sequencing: Effecient Processing of Large Sets of Sequencing Reactions," *Proc. Natl. Acad. Sci. USA*, vol. 91, 2245–2249 (1994).

Lamture, J. et al., "Direct Detection of Nucleic Acid Hybridization on the Surface of a Charge Coupled Device," *Nucleic Acids Research*, vol. 22, No. 11, 2121–2125 (1994).

Mauro, J. et al., "Fiber-Optic Fluorometric Sensing of Polymerase Chain Reaction-Amplified DNA Using an Immobilized DNA Capture Protein," *Analytical Biochemistry*, vol. 235, 61–72 (1996).

Maxam, A. and Gilbert, W., "A New Method for Sequencing DNA," *Proc. Natl. Acad. Sci. USA*, vol. 74, No. 2, 560–564 (1977).

Metzker, M. et al., "Termination of DNA Synthesis by Novel 3'-Modified-Deoxyribonucleoside 5'-Triphosphates," *Nucleic Acids Res.*, vol. 22, No. 20, 4259–4267 (1994). Nikiforov, T. et al., "Genetic Bit Analysis: A Solid Phase Method for Typing Single Nucleotide Polymorphisms," *Nucleic Acids Res.*, vol. 22, No. 20, 4167–4175 (1994). Riccelli, P. and Benight, A., "Tetramethylammonium Does not Universally Neutralize Sequence Dependent DNA Stability," *Nucleic Acids Res.*, vol. 21, No. 16, 3786–3788 (1993).

Rosenthal, A. et al., "Large-Scale Production of DNA Sequencing Templates by Microtitre Format PCR," *Nucleic Acids Research*, vol. 21, No. 1, 173–174 (1993).

Sanger, F. et al., "DNA Sequencing with Chain-Terminating Inhibitors," *Proc. Natl. Acad. Sci. USA*, vol. 74, No. 12, 5463–5467 (1977).

Shoffner, M. et al., "Chip PCR. I. Surface Passivation of Microfabricated Silicon–Glass Chips for PCR," *Nucleic Acids Research*, vol. 24, No. 2, 375–379 (1996).

Sokolov, B., "Primer Extension Technique for the Detection of Single Nucleotide in Genomic DNA," *Nucleic Acids Res.*, vol. 18, No. 12, 3671 (1989).

Strezoska, Z. et al., "DNA Sequencing by Hybridization: 100 Bases Read by a Non-Gel-Based Method," *Proc. Natl. Acad. Sci. USA*, vol. 88, 10089–10093 (1991).

Syvänen, A. et al., "Convenient and Quantitative Determination of the Frequency of a Mutant Allele Using Solid-Phase Minisequencing: Application to Aspartylglucosaminuria in Finland," *Genomics*, vol. 12, 590–595 (1992).

Versalovic, J. et al., "Distribution of Repetitive DNA Sequences in Eubacteria and Application to Fingerprinting of Bacterial Genomes," *Nucleic Acid Res.*, vol. 19, No. 24, 6823–6831 (1991).

Warren S., "The Expanding World of Trinucleotide Repeats," *Science*, vol. 271, 1374–1375 (1996).

Wilding, P. et al., "Manipulation and Flow of Biological Fluids in Straight Channels Micromachined in Silicon," *Clin. Chem.*, vol. 40, No. 1, 43–47 (1994).

Williams, J., et al., "Studies of Oligonucleotide Interactions by Hybridisation to Arrays: The Influence of Dangling Ends on Duplex Yield," *Nucleic Acids Res.*, vol. 22, No. 8, 1365–1367 (1994).

Woolley, A. and Mathies, R., "Ultra-High-Speed DNA Fragment Seperations Using Microfabricated Capillary Array Electrophoresis Chips," *Proc. Natl. Acad. Sci. USA*, vol. 91, 11348–11352 (1994).

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#### [57] ABSTRACT

An iterative and regenerative method for sequencing DNA is described. This method sequences DNA in discrete intervals starting at one end of a double stranded DNA segment. This method overcomes problems inherent in other sequencing methods, including the need for gel resolution of DNA fragments and the generation of artifacts caused by single-stranded DNA secondary structures. A particular advantage of this invention is that it can create offset collections of DNA segments and sequence the segments in parallel to provide continuous sequence information over long intervals. This method is also suitable for automation and multiplex automation to sequence large sets of segments.

#### 118 Claims, 9 Drawing Sheets



# FIG. 1

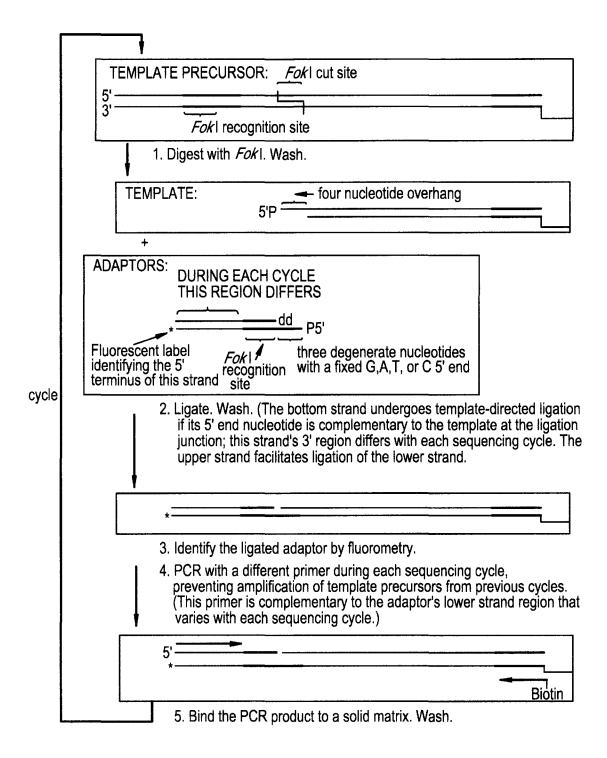
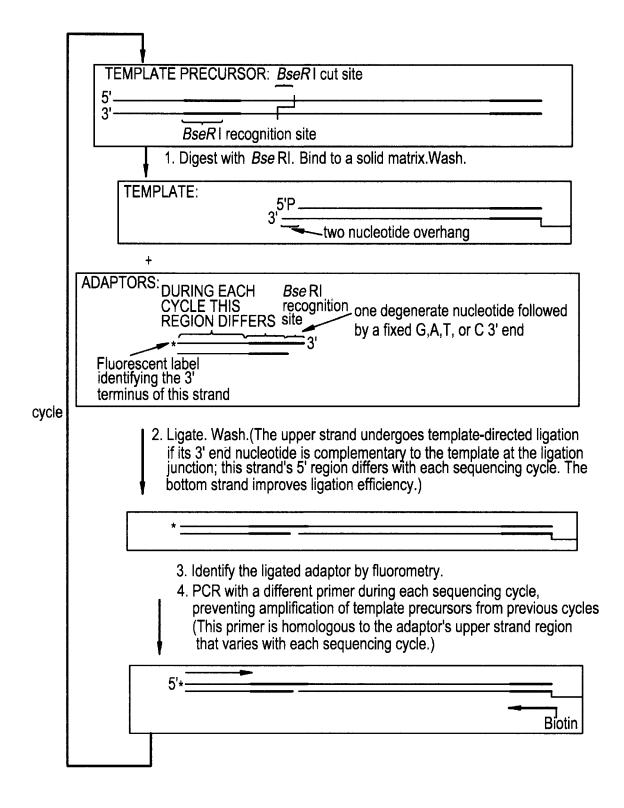




FIG. 2



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