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(12) **United States Patent**
Efcavitch et al.(10) **Patent No.:** **US 9,279,149 B2**
(45) **Date of Patent:** ***Mar. 8, 2016**(54) **METHODS AND APPARATUS FOR SYNTHESIZING NUCLEIC ACIDS**(71) Applicant: **Molecular Assemblies, Inc.**, San Carlos, CA (US)(72) Inventors: **J. William Efcavitch**, San Carlos, CA (US); **Suhaib Siddiqi**, Burlington, MA (US)(73) Assignee: **Molecular Assemblies, Inc.**, San Diego, CA (US)

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(51) **Int. Cl.****C12Q 1/68** (2006.01)**C12P 19/34** (2006.01)(52) **U.S. Cl.**CPC **C12Q 1/6844** (2013.01); **C12P 19/34** (2013.01); **C12Q 1/6806** (2013.01); **B01J 2219/00596** (2013.01); **B01J 2219/00722** (2013.01)(58) **Field of Classification Search**

CPC C12Q 1/6844

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See application file for complete search history.

(56)

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(57)

ABSTRACT

The invention provides improved methods for synthesizing polynucleotides, such as DNA and RNA, using enzymes and specially designed nucleotide analogs. Using the methods of the invention, specific sequences of polynucleotides can be synthesized de novo, base by base, in an aqueous environment, without the use of a nucleic acid template. Because the nucleotide analogs have an unmodified 3' OH, i.e., as found in "natural" deoxyribose and ribose molecules, the analogs result in natural polynucleotides suitable for incorporation into biological systems.

47 Claims, 16 Drawing Sheets