



US006936433B2

(12) **United States Patent**
Akeson et al.

(10) **Patent No.:** **US 6,936,433 B2**
(45) **Date of Patent:** **Aug. 30, 2005**

(54) **METHODS AND DEVICES FOR CHARACTERIZING DUPLEX NUCLEIC ACID MOLECULES**

WO WO 98/35012 * 8/1998
WO WO00/28312 5/2000
WO WO 01/18251 3/2001
WO WO 01/42782 6/2001

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(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 263 days.

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(21) Appl. No.: **09/990,102**

Primary Examiner—Louise N. Leary

(22) Filed: **Nov. 21, 2001**

(74) *Attorney, Agent, or Firm*—Bret E. Field, Bozicevic, Field & Francis LLP

(65) **Prior Publication Data**

US 2003/0099951 A1 May 29, 2003

(57) **ABSTRACT**

Related U.S. Application Data

(60) Provisional application No. 60/253,393, filed on Nov. 27, 2000.

Methods and devices are provided for characterizing a duplex nucleic acid, e.g., a duplex DNA molecule. In the subject methods, a fluid conducting medium that includes a duplex nucleic acid molecule is contacted with a nanopore under the influence of an applied electric field and the resulting changes in current through the nanopore caused by the duplex nucleic acid molecule are monitored. The observed changes in current through the nanopore are then employed as a set of data values to characterize the duplex nucleic acid, where the set of data values may be employed in raw form or manipulated, e.g., into a current blockade profile. Also provided are nanopore devices for practicing the subject methods, where the subject nanopore devices are characterized by the presence of an algorithm which directs a processing means to employ monitored changes in current through a nanopore to characterize a duplex nucleic acid molecule responsible for the current changes. The subject methods and devices find use in a variety of applications, including, among other applications, the identification of an analyte duplex DNA molecule in a sample, the specific base sequence at a single nucleotide polymorphism (SNP), and the sequencing of duplex DNA molecules.

(51) **Int. Cl.**⁷ **C12Q 1/37**; C12Q 1/00;
C12Q 1/68; G01N 33/53

(52) **U.S. Cl.** **435/23**; 435/24; 435/6;
435/4; 435/975

(58) **Field of Search** 435/23, 24, 6,
435/4, 975

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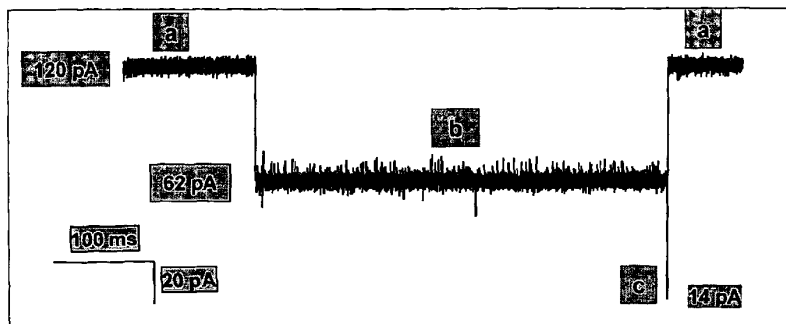
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20 Claims, 18 Drawing Sheets



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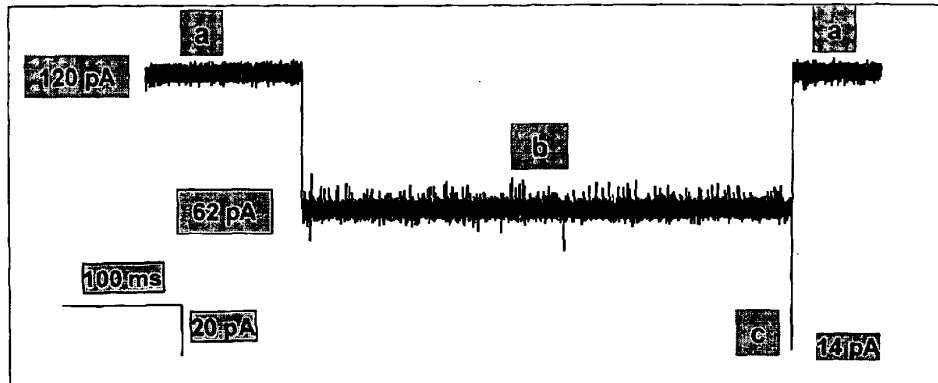


Figure 1

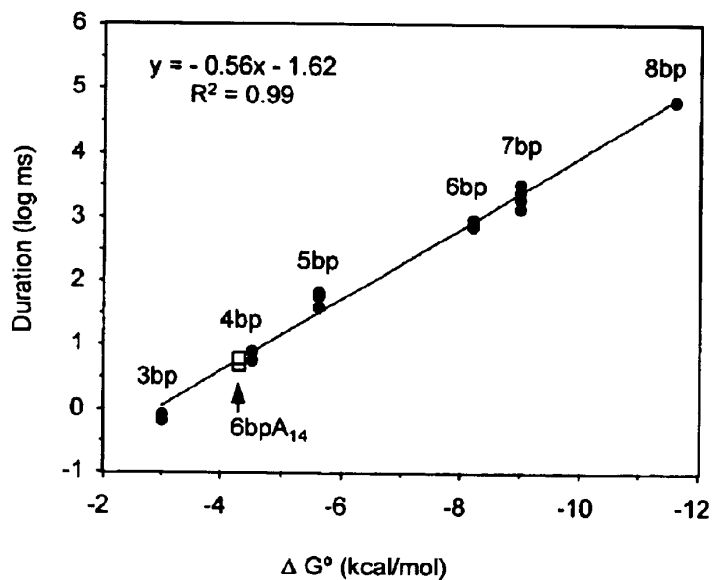


Figure 2

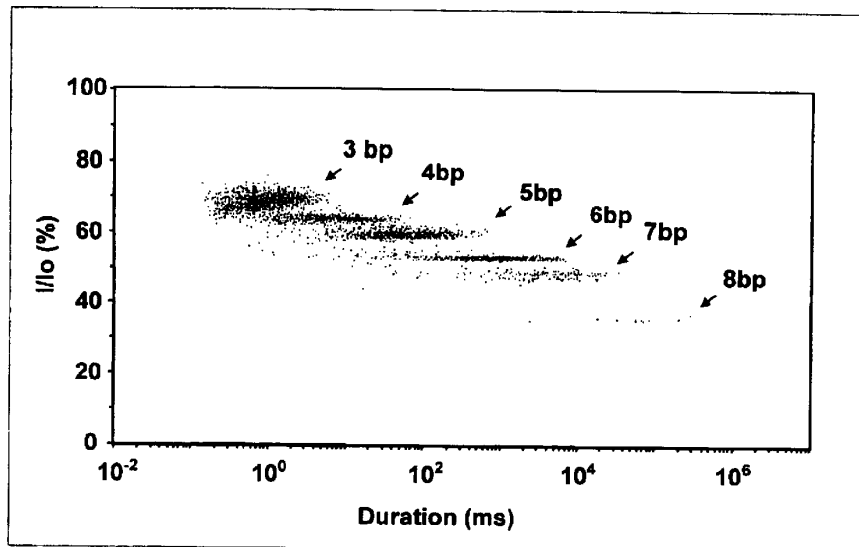


Figure 3a

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