

(12) **United States Patent**  
**Nagalla et al.**(10) **Patent No.:** **US 8,530,175 B2**  
(45) **Date of Patent:** **Sep. 10, 2013**(54) **METHODS FOR DETECTING  
PRE-DIABETES AND DIABETES USING  
DIFFERENTIAL PROTEIN GLYCOSYLATION**(75) Inventors: **Srinivasa R. Nagalla**, Hillsboro, OR  
(US); **Charles T. Roberts**, Portland, OR  
(US)(73) Assignee: **DiabetOmics, LLC**, Beaverton, OR  
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11, 2009.(51) **Int. Cl.**  
**G01N 33/53** (2006.01)(52) **U.S. Cl.**  
USPC ..... **435/7.1; 435/7.2**(58) **Field of Classification Search**  
None  
See application file for complete search history.(56) **References Cited****U.S. PATENT DOCUMENTS**

4,168,146 A 9/1979 Grubb et al.  
4,246,339 A 1/1981 Cole et al.  
4,275,149 A 6/1981 Litman et al.  
4,277,560 A 7/1981 Gray et al.  
4,313,734 A 2/1982 Leuving  
4,366,241 A 12/1982 Tom et al.  
4,371,374 A 2/1983 Cerami et al.  
4,373,932 A 2/1983 Gribnau et al.  
4,435,504 A 3/1984 Zuk et al.  
4,496,654 A 1/1985 Katz et al.  
4,632,901 A 12/1986 Valkirs et al.  
4,703,017 A 10/1987 Campbell et al.  
4,740,468 A 4/1988 Weng et al.  
4,743,560 A 5/1988 Campbell et al.  
4,770,853 A 9/1988 Bernstein  
4,775,636 A 10/1988 Moeremans et al.  
4,806,311 A 2/1989 Greenquist  
4,806,312 A 2/1989 Greenquist  
4,812,293 A 3/1989 McLaurin et al.  
4,855,240 A 8/1989 Rosenstein et al.  
4,857,453 A 8/1989 Ullman et al.  
4,861,711 A 8/1989 Friesen et al.  
4,920,046 A 4/1990 McFarland et al.  
4,943,522 A 7/1990 Eisinger et al.  
4,945,042 A 7/1990 Geiger et al.  
4,954,452 A 9/1990 Yost et al.  
5,001,049 A 3/1991 Klein et al.  
5,073,484 A 12/1991 Swanson et al.

5,075,078 A 12/1991 Osikowicz et al.  
5,120,643 A 6/1992 Ching et al.  
5,126,241 A 6/1992 Schenk  
5,229,073 A 7/1993 Luo et al.  
5,279,935 A 1/1994 Nycz  
5,424,193 A 6/1995 Pronovost et al.  
5,451,504 A 9/1995 Fitzpatrick et al.  
5,451,507 A 9/1995 Skold et al.  
5,591,645 A 1/1997 Rosenstein  
5,712,172 A 1/1998 Huang et al.  
5,798,273 A 8/1998 Shuler et al.  
6,001,658 A 12/1999 Fredrickson  
6,258,548 B1 7/2001 Buck  
6,283,761 B1 9/2001 Joao  
6,368,876 B1 4/2002 Huang et al.  
6,555,390 B2 4/2003 Chandler  
6,656,744 B2 12/2003 Pronovost et al.  
6,699,722 B2 3/2004 Bauer et al.  
6,716,592 B1 4/2004 Rademacher et al.  
7,183,118 B2 2/2007 Aebersold et al.  
8,163,502 B2 4/2012 Denny et al.  
2003/0049857 A1 3/2003 Chan  
2003/0191378 A1 10/2003 Davis et al.

(Continued)

**FOREIGN PATENT DOCUMENTS**

EP 0296724 A2 12/1988  
EP 0299428 A2 1/1989

(Continued)

**OTHER PUBLICATIONS**

Poland, Dennis C.W. et al., "Increased alpha 3-Fucosylation of alpha 1-acid Glycoprotein in Type I Diabetic Patients is Related to Vascular Function," Glycoconjugate Journal, 2001, vol. 18, pp. 261-268.  
Carlsson, Jenny et al., "Detection of Global Glycosylation Changes of Serum Proteins in Type I Diabetes Using a Lectin Panel and Multivariate Data Analysis," Talanta, 2008, vol. 76, pp. 333-337.  
Buse, Maria G. et al., "Enhanced O-GlcNAc Protein Modification is Associated with Insulin Resistance in GLUT1-Overexpressing Muscles," Am J Physiol Endocrinol Metab, 2002, vol. 283, pp. E241-E250.  
Higai, Koji et al., "Altered Glycosylation of alpha1-acid Glycoprotein in Patients with Inflammation and Diabetes Mellitus," Clinica Chimica Acta, 2003, vol. 329, pp. 117-125.  
Wang, Linjie et al., "Concanavalin A-captured Glycoproteins in Healthy Human Urine," Molecular & Cellular Proteomics 5.3, 2006, pp. 560-562.

(Continued)

*Primary Examiner* — Jacob Cheu(74) *Attorney, Agent, or Firm* — Schwabe, Williamson & Wyatt(57) **ABSTRACT**

Methods for identifying individuals who are not yet diabetic (pre-diabetic), but who are at significant risk of developing diabetes, such as type 2 diabetes, are disclosed herein. Methods are also provided for the identification of diabetic subjects. Also disclosed are methods for identifying individuals with diabetic complications. The methods include the identification of an overall glycosylation profile of proteins in a biological fluid, such as saliva, urine, or serum. In some examples, the methods include determining the amount of one or more protein in a biological fluid or determining the glycosylation pattern of one or more proteins in a biological fluid.

**14 Claims, 7 Drawing Sheets**