



US005683899A

United States Patent [19]
Stuart

[11] **Patent Number:** **5,683,899**
[45] **Date of Patent:** ***Nov. 4, 1997**

[54] **METHODS AND COMPOSITIONS FOR COMBINATORIAL-BASED DISCOVERY OF NEW MULTIMERIC MOLECULES**

[75] Inventor: **W. Dorsey Stuart, Kaneche, Hi.**

[73] Assignee: **University of Hawaii, Honolulu, Hi.**

[*] Notice: The term of this patent shall not extend beyond the expiration date of Pat. No. 5,643,745.

[21] Appl. No.: **678,462**

[22] Filed: **Jul. 9, 1996**

Related U.S. Application Data

[63] Continuation-in-part of Ser. No. 191,337, Feb. 3, 1994.

[51] Int. Cl.⁶ **C12N 15/04; C12N 5/12**

[52] U.S. Cl. **435/172.2; 435/254.4**

[58] Field of Search **435/172.3, 172.2, 435/6, 7.1, 7.2, 7.21, 254.4, 254.11**

[56] **References Cited**

U.S. PATENT DOCUMENTS

4,486,533	12/1984	Lambowitz	435/172.3
4,727,037	2/1988	Ring	436/548
4,816,405	3/1989	Yelton et al.	435/243
4,880,734	11/1989	Burke et al.	435/69.1
4,885,249	12/1989	Buxton et al.	435/172.3
4,935,349	6/1990	McKnight et al.	435/69.5
5,177,193	1/1993	Boime et al.	530/397
5,364,770	11/1994	Berka et al.	435/69.1

FOREIGN PATENT DOCUMENTS

552569	7/1993	European Pat. Off.	.
WO 95/21263	2/1995	WIPO	.

OTHER PUBLICATIONS

Buczynski, S. et al., *Fungal Genetics Newsletter* (1995 Suppl.) 42A.
 Carattoli, A. et al., *Proc. Natl. Acad. Sci. USA* (1995)92:6612-6616.
 Dalbey, R.E. et al., *TIBS* (1992) 17:474-478.
 Dales et al., *J. Gen. Microbiol.* (1983) 129:3637-3642.
 Downey et al., *Mol. Cell. Biochem.* (1984)59:155-163.
 Kato, E. et al., *Fungal Genetics Newsletter* (1995 Suppl.) 42A.
 Koo, K. and Stuart, W.D., *Genome* (1991)34:644-651.
 MacKenzie, D.A. et al., *J. Gen. Microbiol.* (1993)139:2295-2307.
 Nakano, E.T. et al., *Fungal Genetics Newsletter* (1995 Suppl.) 40:54.
 Payton et al., *J. Bact.* (1977) 129:1222-1226.
 Peberdy, J.F., *Trends in BioTechnology* (1994)12:50-57.
 Perkins et al., "Chromosomal Loci of *Neurospora crassa*", *Microbiological Reviews* (1982) 46:426-570, at 478.
 Stuart, W.D. et al., *Genome* (1988) 30:198-203.
 Yamashita, R.A. et al., *Fungal Genetics Newsletter* (1995 Suppl.) 42A.

Primary Examiner—James Ketter
Attorney, Agent, or Firm—Morrison & Foerster LLP

[57] **ABSTRACT**

The present invention provides methods and compositions for producing and screening combinatorial libraries of multimeric proteins. The present invention relies on the use of filamentous fungal heterokaryons that are produced using two or more parent strains into which a population of DNA molecules encoding variants of a multimeric protein have been introduced.

13 Claims, No Drawings