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1 August 1996 (01.08.96)

(54) Title: SOLUBLE SINGLE CHAIN T CELL RECEPTORS

(57) Abstract

A fusion protein, comprising a carrier protein connected by a peptide tether to a single chain T cell receptor molecule, consisting of a Vα segment linked to a Vβ segment of the T cell receptor, is disclosed. Also disclosed is a soluble single chain T cell receptor molecule with a conformation that is essentially functionally indistinguishable, based upon reactivity to clonotype-specific antibodies, from that appearing on the surface of T cells. The invention also concerns nucleic acid fragments encoding the fusion protein, expression vectors comprising a nucleic acid fragment encoding the fusion protein, host cells containing such expression vectors, and antibodies to the single chain T cell receptor or to the fusion protein. The invention further pertains to methods of isolating and purifying the fusion proteins, as well as isolating and purifying soluble, single chain T cell receptors. In addition the invention pertains to various uses of soluble TCR fusion protein and isolated single chain TCR. The proteins can be used in molecular assays designed to measure their binding to ligands, including MHC/HLA-peptide antigen complexes or TCR-specific antibodies. Such assays are useful for the detection of agents that block the TCR-ligand interaction. The soluble TCR proteins can also be used to immunize animals, including humans, to produce TCR-specific antibodies. In addition, either in their native or denatured conformation the proteins can be used to vaccinate animals, including humans, in order to suppress the immune response of T cells bearing TCR that share antigenic epitopes with the vaccinating protein.

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INTERNATIONAL SEARCH REPORT

International Application No
PCT/US 95/13770

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 C12N15/62 C07K14/725 C12P21/02 C07K16/28 G01N33/68
A61K38/17

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 C12N C07K A61K G01N C12P

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO,A,91 18019 (E.R. SQUIBB & SONS, INC. & DANA FARBER CANCER INSTITUTE) 28 November 1991 see examples see claims ---	8-10,34
X	THE FASEB JOURNAL, vol. 6, no. 1, 1 January 1992, BETHESDA, MD, USA, page A351 XP002002795 J. NOVOTNY ET AL.: "A soluble single chain T cell receptor fragment endowed with antigen-combining properties." see abstract 2022 --- -/--	8-10

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

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2

Date of the actual completion of the international search	Date of mailing of the international search report
10 May 1996	17.06.96
Name and mailing address of the ISA European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax (+31-70) 340-3016	Authorized officer Nooij, F

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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE USA, vol. 88, no. 19, 1 October 1991, WASHINGTON, DC, USA, pages 8646-8650, XP002002796 J. NOVOTNY ET AL.: "A soluble, single-chain T cell receptor fragment endowed with antigen-combining properties." cited in the application see abstract see page 8647, right-hand column, line 26 - line 32 ---	8-10,21, 23,34
X	PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE USA, vol. 89, no. 10, 15 May 1992, WASHINGTON, DC, USA, pages 4759-4763, XP002002797 W. SOO HOO ET AL.: "Characterization of a single-chain T-cell receptor expressed in Escherichia coli." see the whole document ---	8-10,21, 24,33,34
X	WO,A,93 22332 (BOARD OF REGENTS, THE UNIVERSITY OF TEXAS SYSTEM) 11 November 1993 see page 29, line 33 - page 32, line 20 see seq. id. nos. 7,8 see page 44, line 15 - line 24 see examples 1-4 see claims 1-41 ---	8-10
X	WO,A,92 01715 (THE BOARD OF TRUSTEES OF THE LELAND STANFORD JUNIOR UNIVERSITY) 6 February 1992 see "experimental" see claims ---	8-10
A	NATURE, vol. 321, 15 May 1986, LONDON, GB, pages 219-226, XP002002798 P. FINK ET AL.: "Correlations between T-cell specificity and the structure of the antigen receptor." cited in the application see figure 3 --- -/--	1-35

2

Form PCT/ISA/210 (continuation of second sheet) (July 1992)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS, vol. 201, no. 3, 30 June 1994, DULUTH, MN, USA, pages 1502-1509, XP002002799 D. LAKE ET AL.: "Construction and serological characterization of a recombinant human single chain T cell receptor." see the whole document ---	8-10
P,X	J. NEWCOMB ET AL.: "Demonstration of soluble T cell receptor function using proliferation-based assays. In: THE 9TH INTERNATIONAL CONGRESS OF IMMUNOLOGY. San Francisco, July 23-29, 1995. Abstract book." 1995 , SAN FRANCISCO, CA, USA XP002002801 see abstract 2616 ---	8-10
P,X	M. JESSON ET AL.: "Immunogenicity of recombinant, soluble T cell receptors in syngeneic mice. In: THE 9TH INTERNATIONAL CONGRESS OF IMMUNOLOGY. San Francisco, July 23-29, 1995. Abstract book." 1995 , SAN FRANCISCO, CA, USA XP002002802 see abstract 2617 ---	8-10,33, 34
P,X	THE FASEB JOURNAL, vol. 9, no. 3, 9 March 1995, BETHESDA, MD, USA, page A514 XP002002800 C. WONG ET AL.: "Single chain T cell receptor as a tumor specific vaccine." see abstract 2976 -----	1,8-11, 17,18, 21,23,34

2

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US95/13770

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.: 26,27,33,34
because they relate to subject matter not required to be searched by this Authority, namely:

Remark: Although these claims are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

2. [ ] Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. [ ] As all searchable claims could be searches without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

International Application No
PC1/US 95/13770

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
WO-A-9118019	28-11-91	AU-B-	652376	25-08-94
		AU-B-	8206391	10-12-91
		CA-A-	2047733	16-11-91
		EP-A-	0483351	06-05-92
		JP-T-	5500068	14-01-93
WO-A-9322332	11-11-93	AU-B-	4116793	29-11-93
		CA-A-	2118508	11-11-93
		EP-A-	0640094	01-03-95
WO-A-9201715	06-02-92	NONE		