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(54) Title: HAPLOTYPES OF THE EDG1 GENE

(57) Abstract: Novel genetic variants of the Endothelial Differentiation, Sphingolipid G Protein-Coupled Receptor 1 (EDG1) gene are described. Various genotypes, haplotypes, and haplotype pairs that exist in the general United States population are disclosed for the EDG1 gene. Compositions and methods for haplotyping and/or genotyping the EDG1 gene in an individual are also disclosed. Polynucleotides defined by the haplotypes disclosed herein are also described.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US01/46946

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) :C07H 21/02, 21/04; C12Q 1/68, 1/70; C12P 19/34
US CL :536/22.1, 23.1, 24.3, 24.31; 435/6, 91.1, 91.2
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)

U.S. : 536/22.1, 23.1, 24.3, 24.31; 435/6, 91.1, 91.2

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 practicable, search terms used)

Please See Extra Sheet.

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	HLA et al. An abundant transcript induced in differentiating human endothelial cells encodes a polypeptide with structural similiarities to G-protein-coupled receptors. J. Biol. Chem. 05 June 1990, Vol. 265, No. 16, pages 9308-9313, see entire document.	1, 2, 6, 7
Y	ANet al. Identification of cDNAs encoding two G protein-coupled receptors for lysosphingolipids. FEBS Lett. 1997, Vol. 417, pages 279-282, see entire document.	1, 2, 6, 7
Y	US 5,521,301 A (WALLACE et al) 28 May 1996, see entire document.	1, 2, 6, 7
Y	US 5,851,762 A (SIMONS) 22 December 1998, see entire document.	1, 2, 6, 7

[X] Further documents are listed in the continuation of Box C. [ ] See patent family annex.

*	Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"A"	document defining the general state of the art which is not considered to be of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"E"	earlier document published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"L"	document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family
"O"	document referring to an oral disclosure, use, exhibition or other means		
"P"	document published prior to the international filing date but later than the priority date claimed		

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Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703) 305-3230	Authorized officer JEFFREY FREDMAN Telephone No. (703) 308-0196

Form PCT/ISA/210 (second sheet) (July 1998)★

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US01/46946

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

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

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

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:

Please See Extra Sheet.

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 searched 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.:
1, 2, 6, 7

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.
PCT/US01/46946

B. FIELDS SEARCHED
Electronic data bases consulted (Name of data base and where practicable terms used):

EAST, MEDLINE, BIOSIS, CAPLUS, REGISTRY
search terms: edg1 endothelial, g, protein, coupled, receptor, haplotype, genotype, sequencing, polymorphism, allele, variant, alteration, insertion, deletion, transposition, substitution

BOX II. OBSERVATIONS WHERE UNITY OF INVENTION WAS LACKING
This ISA found multiple inventions as follows:

Groups 1-25, claim(s) 1-2, 6, 7, in part, drawn to methods for haplotying EDG1 comprising determining whether the individual has one of the EDG1 haplotypes shown in the tables. The first Group examined will be Haplotype 1. If Applicant elects additional groups, Applicant is requested to identify the haplotype or haplotype pair elected, whichever Group is elected.

Groups 26-35, claim(s) 3-5, in part drawn to a method for genotyping the EDG1 gene.

Groups 36-50, claim(s) 8-9, in part drawn to a method for predicting a haplotype pair for the EDG1 gene by identifying a EDG1 genotype for the individual at two or more polymorphic sites PS1-9.

Groups 51-75, claim(s) 10-11, in part drawn to a method for identifying an association between a trait and a haplotype between one of the 25 haplotypes and haplotype pairs of EDG1 gene.

Groups 76-119, claim(s) 12-16, in part, drawn to a composition comprising at least one genotping oligonucleotide for detecting a polymorphism in the EDG1 gene. If one of these groups is selected, Applicant must elect one SEQ ID NO for examination.

Groups 120-129, claims 17, drawn to a kit comprising a set of oligonucleotides designed to genotype each of the polymoprhic sites.

Groups 130-139, claims 18, 19, 22, 23, 26, in part, drawn to a polynucleotide which is a polymorphic variant of a reference sequence for EDG1 gene or a fragment thereof.

Groups 140-149, claim(s) 20-21 and 24-25, in part drawn to a recombinant nonhuman organisms comprising one of the 24 haplotypes or haplotype pairs respectively.

Groups 150-152, claim(s) 27 and 30, in part drawn to a polypeptide comprising an amino acid sequence which is a polymorphic variants of a reference sequence for the EDG1 protein or a fragment thereof.

Groups 153-155, claim(s) 28, in part drawn to an antibody which binds to a polypeptides of Claim 30.

Groups 156-165, claim(s) 29, in part drawn to a method for screening for drugs targeting the EDG1 polypeptide.

Groups 166-180, claim(s) 31, in part drawn to a computer system comprising polymorphism data wherein the data comprises the haplotypes and haplotype pairs shown in the tables.

Groups 181-190, claim(s) 32, in part, drawn to a genome anthologies comprising EDG1 isogenes having any one of the haploytpes or haplotype pairs of the tables.

The products claimed in Claims 12-17 include fragments of variant sequences, and the claims simply require the presence of a single polymorphic site in the coding sequence, so that a random hexamer mixture, commercially available, would anticipate the claims. Accordingly, the claims are sufficiently broad so as to encompass nucleic acid fragments taught in the art. Further, the amino acid fragment claims such as claim 30 simply require the fragment comprise one variant amino acid such as aspartic acid. Since the fragment could be a single amino acid, aspartic acid was known prior to the current application. As the nucleic acid and protein fragment products do not represent a contribution over the prior art, the claims lack a special technical feature that is the same as or that corresponds to a special technical feature of the other claimed inventions. Thus, there is no special technical

feature linking the recited Groups, as would be necessary to fulfill the requirement for unity of invention.

It is also noted that each of the present claims has been presented in improper Markush format, as distinct products and distinct methods are improperly joined in the claims. With respect to the claims, each polymorphic site and each molecule containing said polymorphic site is structurally and functionally distinct from and has a different special technical feature than each other polymorphic site and molecules containing said site. The chemical structure of each polymorphism and of each molecule containing the same differ from each other. For example, a polynucleotide comprising PS1 is chemically, structurally, and functionally different from a molecule comprising PS3. As the products and methods encompassed by the claims do not share a special technical feature, the distinct products and methods may not properly be presented in the alternative. Accordingly, the claims have been separated into a number of groups corresponding to the number of different inventions encompassed by the claims, and the claims will be examined only as they read upon the invention of the elected group. For the same reasons, the remainder of the claims have been separated in a number of groups corresponding to the number of different inventions encompassed thereby.

With particular respect to the haplotype and genotyping claims, it is noted that the haplotypes and genotypes encompassed by these claims are also distinct from each other and from the single polymorphisms recited. For example, a molecule of haplotype 1, comprising a particular combination of polymorphisms, differs chemically, structurally, and functionally from a molecule of haplotype 2 and from a molecule comprising a single polymorphism (e.g., PS1). The special technical feature of each haplotype or genotype is the combination of polymorphisms contained therein, which feature is lacking from and not shared with each other haplotype or genotype or with, e.g., a molecule comprising any single polymorphism set forth in the claims. Similarly, with respect to the pairs of polymorphism, each combination of polymorphism differs from each other combination and from each of the other combinations discussed above (i.e., haplotypes, genotypes, and single polymorphic sites). Thus, the claims have been separated into a number of groups corresponding to the number of different inventions encompassed thereby, and the claims will be examined only as they read upon the invention of the elected group.

Further polynucleotides, kits, and various compositions, recombinant organisms, polypeptides, antibodies, computer system, and genome anthologies are additionally drawn to multiple, distinct products lacking the same or corresponding special technical features. The nucleic acids are composed of nucleotides and function in , e.g., methods of nucleic acid hybridization or amplification. These groups are directed to different combinations of nucleic acids which are different from one another and may be employed in different methods. The recombinant organisms are complex organisms that are employed in, e.g. animal research methods. Such organisms cannot be employed as, e.g., probes or primers and they differ in both structure and function from the nucleic acids. The polypeptides differ in both structure and function from either the nucleic acids or the transgenic organisms. The polypeptides are composed of amino acids linked by peptide bonds and arranged in a complex combination of alpha helices, beta pleated sheets, hydrophobic and hydrophilic domains. The polypeptides also differ in function, e.g., fusion proteins with an enzymatic functions. The antibodies are composed of amino acids linked by peptide bonds, antibodies are glycosylated and their tertiary structure is unique, where four subunits (2 light chains and 2 heavy chains) associated via disulfide bonds into a Y-shaped symmetric dimer. The antibodies function in immunoassays. Further the computer systems are composed of, e.g., a CPU, a display device, an input device, etc., and function in, e.g., methods of electronic sequence comparison. Accordingly, the products of each of these Groups differ structurally and functionally from each other. As products of different sets of Groups differ from each other in structure, function, and effect, they do not belong to a recognized class of chemical compound, or have both a "common property or activity" and a common structure as would be required to show that the inventions are "of a similar nature".

Further, the methods of each of the method Groups have different objectives and require different process steps. The haplotyping methods of require steps of identifying haplotypes and haplotype pairs to achieve the objectives of haplotyping. The genotyping methods require steps of identifying a single nucleotide on one gene copy to achieve the objective of genotyping. The predictive methods require steps of identifying two polymorphisms in a gene to achieve the objective of "predicting a haplotype pair". The association methods requires steps of comparing frequencies of haplotypes in a population to achieve the objective of "identifying an association between a trait" and a haplotye. The ligand screening methods require steps of assaying for binding activity for candidate agents. In addition to differences in objectives, effects, and method steps, it is again noted that the claims of the present Groups are not directed to the detection or identification of molecules having the same or common special technical feature, for the reasons discussed above.