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(54) **Title:** USE OF ILT6 FOR IMMUNOMODULATION

(57) **Abstract:** The present invention relates to the modulation of the immune response in human beings. In greater detail, the present invention relates to the medical use of ILT6 for modulating the immune response, as well as to pharmaceutical compositions containing ILT6. In a further aspect, the present invention relates to the medical treatment of human beings using ILT6 to modulate their immune response, for example directed against heterologous and/or autologous antigen. Furthermore, the present invention relates to the analysis ILT6 for diagnostic purposes and to diagnostic compositions that can be employed for that analysis.

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New PCT application
Immunomodulation – Medizinische Hochschule Hannover, Dr. T. Witte, Dr. T. Matthias

Immunomodulation

The present invention relates to the modulation of an immune response in human beings. In greater detail, the present invention relates to the medical use of a compound for modulating the immune response, as well as to pharmaceutical compositions containing said compound.

In a further aspect, the present invention relates to the medical treatment of human beings using said compound to modulate their immune response, for example directed against heterologous and/or autologous antigen. Furthermore, the present invention relates to the analysis of said compound for diagnostic purposes and to diagnostic compositions that can be employed for that analysis or for the analysis of antibody cross-reactive with said compound.

State of the art

It is known to modulate an undesired activity of the immune system by generally suppressing the activity of the immune system. As an example, chemotherapy or the medical administration of hormones like cortisone are employed.

Also known are genes encoded by the leucocyte Ig-like receptor cluster (LILR), containing at least thirteen genes encoding Ig-like transcripts (ILT) (Trowsdale et al., *Immunol. Rev.* 181, 20 – 38 (2001)).

ILTs are expressed by a variety of immune cells and it is known that ILT2 and ILT4 bind to HLA I molecules. It has been described by Chang et al., *Nat. Immunol.*, 3: 237 – 243 (2002) that the expression of ILT3 and ILT4 may play a role in the generation of tolerance in T-cells. The regulation of the immune response towards antigen is exerted by ILTs by inhibiting or activating the cytolytic activity (Dietrich et al., *J. Immunol.*, 166: 2514 – 2521 (2001)).

For ILT6, also termed LILRA3 or CD85e, it has been found that this is a soluble protein, i.e. it is not membrane-bound. Furthermore, Torkar et al., *Eur. J. Immunol.*, 30: 3655 – 3662 (2000) have described that individuals lacking the seven 5′exons of the gene encoding ILT6 (deletion of a 6.7 kbp segment) exhibit a presence / absence variability for ILT6.

According to the DNA sequence, ILT6 has a certain homology to the ILT2 gene. The gene for ILT6 is localized on 19q13, a chromosomal region that has been shown to carry risk genes for multiple sclerosis (MS), a common demyelinating disease affecting the central nervous system. MS is characterized by a progressive destruction of the central nervous system by the individual′s immune system, wherein the cellular immune response plays a significant role.

ILT2 is expressed on professional antigen presenting cells (APC) as well as on T-cells and natural killer cells. ILT2 binds to HLA I and acts as a receptor. ILT2 modulates the activity of T-cells and is participating in the induction of immunological tolerance by interacting with regulatory T-cells. For example, ILT2 participates in mediating tolerance of the immune system against mucosal antigens. It is furthermore presumed that ILT2 participates in the maternal / fetal immunotolerance during pregnancy.

Objects of the invention

It is a first object of the present invention to provide a compound which can be used for medical purposes, e.g. in pharmaceutical compositions, to modulate the activity of the immune system of human beings. It is especially desired to provide a compound that

predominantly exerts its effect on the cellular immune response. Furthermore, it is desired to provide a compound that can be used to activate or repress the activity of the immune system. In this respect, it is especially desired to provide a compound acting upon the activity of T-cells that form part of an acute immune response, preferably activated T-cells.

General description of the invention

The present invention achieves the above-mentioned objects by providing ILT6, obtainable as the translation product from the human gene encoding ILT6. Alternatively, functional derivatives of the human ILT6 gene product are provided, obtainable for example by mutation, derivatization, deletion and/or fusion with itself or other peptides.

It has been found by the present inventors that ILT6, which is a soluble ILT, modulates the cellular immune response in a dose dependent manner. In detail, it has been found that low concentrations of ILT6, for example below 0.01 µg/mL, activate the proliferation of lymphocytes in an *in vitro* format, whereas concentrations higher than 0.01 µg/mL ILT6 inhibit the proliferation of lymphocytes *in vitro*.

ILT6 can be produced by known procaryotic and eucaryotic expression systems for heterologous protein expression from a DNA sequence derived from the human ILT6 gene. When expressing ILT6 in mammalian cells, the natural organization of the ILT6 gene including its intron sequences may be used, whereas for procaryotic expression systems or expression systems using yeasts or fungi, it is preferred to employ the DNA sequence devoid of intron sequences e.g. a DNA sequence derived from the cDNA of the processed transcript from the ILT6 gene.

In accordance with the above-mentioned objects of the invention, there is provided the human ILT6 gene product as well as its use for medical applications or as an active ingredient in pharmaceutical compositions suitable for indications of undesired immune responses like an autoimmune disease, e.g. MS or Sjögren´s syndrome. Furthermore there is provided a diagnostic assay testing for the concentration of functionally active ILT6 in human beings, preferably in patients suffering from an autoimmune disease, e.g. MS or Sjögren´s syndrome. In a further embodiment of the invention, there is provided a diagnostic assay testing for the

concentration of antibody cross-reactive with ILT6, which antibody is present in patients, preferably in patients suffering from an autoimmune disease, e.g. MS or Sjögren's syndrome.

The present inventors have found that in a group of 751 patients with MS, homozygous ILT6 deficiency could be detected in 7.3%, but only in 3.8 % of a control group, showing a statistically significant association with multiple sclerosis, especially the remitting relapsing MS, but not with primary progressive MS or secondary progressive MS. In controls, the distribution of homozygous and heterozygous ILT6 deletion did not differ from the Hardy-Weinberg equilibrium.

As many autoimmune diseases are polygenic, the deficiency of ILT6 in MS patients may be one of several contributing factors. The present inventors assume that the deficiency of ILT6 alone is not a prerequisite for the development of MS, as can be seen from its relation with remitting relapsing MS only. However, the differences of the association of ILT6 deficiency with RRMS (8.0%) to PPMS (7.0%) and SPMS (5.8%) are not statistically significant and may be due to the small sample sizes (RRMS, n = 451; PPMS, n = 129; SPMS, , n = 154). Accordingly, it cannot at present be concluded that ILT6 deficiency is characteristic for a specific subgroup of MS.

Although the mechanism for the action of ILT6 is not known, it is assumed that ILT6 may be acting as an agonist for a related ILT that binds to the same ligand, especially to MHC I. In the alternative, ILT6 could be acting as a soluble mediator between an antigen and cells of the immune system.

It could be shown that ILT6 predominantly exerts its immunomodulating effect on T-cells that have been activated. For example, monocytes and macrophages activated by interferon γ are immunomodulated by the presence of ILT6, however, without significantly influencing resting immune cells, e.g. resting monocytes, B-cells, T-cells or PBL (peripherial blood lymphocytes). For the purposes of immunomodulating the activity of activated immune cells, the natural translation product of the human ILT6 gene sequence can be used, for example expressed in animal cell culture from the structural gene for human ILT6. In the alternative, an identical translation product can be obtained from translation of a gene sequence in microorganisms which is derived from the human ILT6 gene sequence from which the

intermittent intron sequences have deleted, optionally, signal sequences which are active in microorganisms used for the production of ILT6 protein may be added.

Furthermore, functional derivatives of ILT6 can be used for the purposes of this invention. Such functional derivatives comprise for example fusion proteins of different proteins or ILT6 itself with one or more regions of the natural ILT6 protein, and exchanges, deletions or substitutions of single amino acids or amino acid regions within the ILT6 protein. Examples for fusions with other peptides are tags useful for purification like the poly-histidine tag and antigenic sites which are detectable by known antibodies. Cloning and expression of functional derivatives of human ILT6 protein can be performed according to standard practice by a person skilled in the art without undue experimentation. The functional activity of ILT6 and its derivatives can be determined by a person skilled in the art by testing the immunomodulating characteristics of the protein, e.g. in a mixed lymphocyte assay. An example for such a functional test is given in example 3.

It is a specific advantage of ILT6 that it immunomodulates an active immune response i.e. an acute immune response by activated immune cells. Therefore, ILT6 can be employed for medical use, for example as an active component in pharmaceutical preparations for modulating an active immune response, for example for influencing the course of an acute phase of an immune response or a permanent immune response. In its preferred embodiment, the present invention employs ILT6 for medical use essentially without influencing resting immune cells. As an example, ILT6 can be used for treating immune responses due to for example the graft versus host disease or the host versus graft disease in transplant patients, or autoimmune diseases. Examples for autoimmune diseases are multiple sclerosis and Sjögren's syndrome, wherein ILT6 can be used to suppress the immune reaction, preferably during acute phases of the disease.

The administration of ILT6 for medical purposes can be obtained by injection, whether i.m., s.c., or i.v., or orally, using suitable formulations.

It has been found that the presence and concentration of ILT6 in body fluid, e.g. blood samples obtained from a human are in relation to the state of the human's immune system, e.g. the presence of an acute immune response.

Accordingly, the present invention in a further aspect provides a diagnostic assay to determine the concentration and/or activity of ILT6 in a sample of body fluid. As a result of the determination of the concentration and/or activity of ILT6, the state of the immune system of a human being can be determined, for example the presence or absence of an acute immune response. When relating the presence or absence of an acute immune response as determined by one of the analytical methods, to further symptoms or indications of the individual, the analytical methods can be used to determine the state or activity of an autoimmune disease, for example MS or Sjögren´s syndrome.

In one embodiment, a diagnostic test relating to ILT6 is the analysis of the concentration of ILT6 in a sample of body fluid, e.g. blood, by an antibody based assay. One example for an antibody based assay is an ELISA format using at least one monoclonal or polyclonal antibody specific for ILT6 and a suitable detection system. The ELISA format may be non-competitive or competitive, for example using ILT6 protein expressed in eucaryotic or procaryotic cells as the competitor for ILT6 present in the sample of body fluid. Another format is a dot blot or Western blot of a sample of body fluid using the antibody directed against ILT6 for detecting ILT6 or cross-reactive derivatives thereof.

Antibody specific for ILT6 can be raised by immunization of a variety of laboratory animals, for example mice, rats and rabbits to obtain serum containing polyclonal antibody. Monoclonal antibody is obtainable by the hybridoma technique, using the fusion of spleen cells isolated from immunized laboratory animals with myeloma cells. A detailed description of production methods for polyclonal and monoclonal antibody is available from "Methods in Enzymology" and "Molecular Cloning, A Laboratory Manual by Sambrook, Maniatis, Fritsch, Cold Spring Harbour".

In consideration of possible immune reactions directed against ILT6 in a human being suffering from an autoimmune disease or another undesired immune response and in view of possible immune reactions against ILT6 administered to a patient during a medical treatment, diagnostic methods and diagnostic compositions for the determination of such immune reactions against ILT6 are within the scope of the present invention.

Accordingly, in a further aspect the present invention provides for a diagnostic test for auto-antibodies directed against ILT6 and for antibodies directed against ILT6 administered to

patients who do not express ILT6. In this respect, ILT6 or functional derivatives thereof are provided for use in an immunological assay to determine the presence and concentration of antibody in a sample of body fluid cross-reactive with ILT6. As described above, ILT6 and its functional derivatives may be produced by heterologous gene expression or, alternatively, synthetically by peptide synthesis, the derivative preferably comprising the epitopes cross-reactive with auto-antibody or antibody directed against ILT6 only or repeatedly.

The diagnostic test may be in the format of an ELISA, blot or any other immunological assay using ILT6 or an immunological equivalent thereof as the capture protein to specifically adsorb antibody cross-reactive with ILT6.

Detailed description of the invention

The invention is now described in greater detail with relation to the accompanying figures, wherein

- Figure 1 is a graph showing the inhibition and stimulation, respectively, of peripheral blood mononuclear cells depending on the concentration of ILT6.

For Sjögren's syndrome, ILT6 deficiency was present in 9% of patients, which is a statistically significant association with the disorder. In contrast, the ILT6 deficiency is not associated with SLE and scleroderma.

Example 1: cloning of the human ILT6 gene

Genomic DNA was extracted from peripheral whole blood using the QiaAmp DNA Minikit (Qiagen, Hilden, Germany). Using PCR primers 5'CCC CCT GGA GCT CGT GG 3' (Seq. ID No. 1) and 5'GAC AGC AGA TTC TAA AAC AGT G 3' (Seq. ID No. 2), the complete ILT6 gene comprising 1150 base pairs was amplified in a in the PCR reaction using 10 pmoles of each primer, 50 ng genomic DNA in 1 x PCR buffer containing 1.5 mM $MgCl_2$, 200 µM dNTPs, 2.5 units Taq polymerase according to the manufacturer's instructions.

Thermocycling used 95 °C for 15 minutes, 30 cycles of 94 °C for 45 seconds, 64 °C for 55 seconds and 72 °C for 55 seconds, and a final extension at 72 °C for 10 minutes. Products

were separated by electrophoresis (1.5% agarose), staining which ethidium bromide and detection under UV.

For sequencing the PCR products were cloned into the PCR 2.1 vector using the TA cloning kit (Invitrogen, Karlsruhe).

Statistical analysis was performed with Fisher's exact test, regarding only associations below 0.05 as significant.

Example 2: Production of ILT6

For production of ILT6 protein, the cDNA encoding human ILT6 was synthesized using RT-PCR on complete RNA isolated from human peripheral whole blood according to known methods, for example using the QIAamp RNA blood mini kit (Qiagen, Hilden, Germany) according to the manufacturer's instructions.

For expression and later purification, the cDNA encoding the complete exons for human ILT6 without interfering intron sequences additionally received a fused sequence encoding a poly-histidine tag in 3'. This is one example for a functional derivative of human ILT6, retaining its biological activity.

For RT-PCR, the following primers were used on cellular RNA: 5' AGG ATC CGC CAT GAC CCC CAT C 3' (Seq. ID No. 3) containing an additional BamH1 site 5'to the start codon and 5'GCG GCC GCT CAA TGA TGA TGA TGA TGA TGC TCA CCA GCC TTG GAG 3' (Seq. ID No. 4), encoding a histidine tag 5'to the stop codon and containing a Not1 site 3' to the stop codon. The resultant gene product is subsequently termed ILT6::poly-His.

Expression of the fusion peptide ILT6::poly-His was cloned into expression vector pBacPak8 (Clontech) using restriction sites BamH1 and Not1 subsequent to ligating the PCR product into vector pCR2.1 (Invitrogen) and sequencing. After transfer into bacculovirus, cultivated SF9 cells were infected. For expression, cells were grown in TNM-FH medium (described in Hink et al., Nature 226: 466-467 (1970) as obtainable from 900 mL Graces medium, combined with 80 mL fetal bovine serum, 3.0 g lactalbumin hydrolysate, 3.0 g Yeastolate, stirred vigorously until powdered ingredients dissolve; if necessary, adjust pH to 6.40 - 6.45

by addition of 1.0 N KOH (increase) or 1.0 N HCl (decrease); if necessary, adjust osmotic pressure to 360 – 380 milliosmols with anhydrous D-glucose (increase) or water (decrease) (obtained from Sigma-Aldrich, product No. T3285) and induced by infection for a period of 5-6 days at 37 °C in a 5% CO_2 atmosphere until plaque formation.

After harvesting cells, the protein was purified from the cellular lysate making use of the His-tag in nickel chelate chromatography. The eluted ILT6::poly-His was eluted from the nickel chelate column and used for immunomodulating experiments.

Example 3: *in vitro* activity of ILT6 on the immune system

Recombinantly produced ILT6::poly-His obtained according to Example 3 was used for modulating the activity of peripheral blood lymphocytes (PBL) in a mixed lymphocyte reaction. The mixed lymphocyte reaction comprised 100,000 PBL/well obtained from a healthy first donor that were stimulated by the addition of 20,000 PBL obtained from a second healthy donor which had been irradiated before their addition.

Recombinant ILT6 obtained according to Example 3 was added to final concentrations between 0.33 µg/mL and 0.0033 µg/mL.

For monitoring the proliferation of the PBL obtained from the first donor, ^{3}H-thymidine was added after 72 hours and the test reactions were harvested after a total of 96 hours. For comparison, protein Ro expressed and purified by the method according to Example 3, replacing the ILT6 gene sequence by the Ro gene sequence, was used.

In Figure 1, the inhibition of proliferation of peripheral blood mononuclear cells in a mixed lymphocyte reaction by addition of ILT6 is shown. The average and standard deviations obtained from experiments with three different donors of stimulating PBL is plotted.

The results demonstrate that high concentrations of ILT6, especially concentrations equal to or above 0.033 µg/mL blocked proliferation of activated PBL, whereas low concentrations of ILT6, especially below 0.0033 µg/mL, were stimulating their proliferation. When using Ro protein in comparative assays, essentially no inhibition of proliferation nor their activation was found.

When examining the morphology of PBL microscopically, it was detected that concentrations of ILT6 inhibiting the proliferation of PBL did not lead to toxic effects to PBL but rather to an arrest of their proliferation.

Example 5: Direct modulation of T-cells by ILT6

For monitoring the intracellular calcium concentrations in T-cells treated with ILT6 at varying concentrations, the following assay was used: PBL or Jurkat T-cells were loaded with 20 µM of the visible light–excitable Ca^{2+} indicator fluoro-h fluorochrome (CAS 121714-22-5 / Glycine, N-[4-[6-[(acetyloxy)methoxy]-2,7- dichloro-3-oxo-3H-xanthen-9-yl]-2-[2-[2- [bis[2-[(acetyloxy)methoxy]-2- oxyethyl]amino]-5- methylphenoxy]ethoxy]phenyl]-N-[2-[(acetyloxy)methoxy]-2-oxyethyl]-, (acetyloxy)methyl ester (obtained from Molecular Probes, c/o Invitrogen, Karlsruhe, Germany, under catalog No. F1241) for 30 min at 37°C, then washed gently three times in PRMI medium and once with PBS (phosphate buffered saline). Finally, calcium flux was induced by addition of ILT6 at various concentrations. As a positive control, 10µM Ionomycin was added to a separate test sample. Intracellular release of calcium was measured at 400 nm in a FACScalibur (Becton Dickinson) fluorescence activated cell sorter.

It was found that presence of ILT6 induces the intracellular liberation of calcium. This demonstrates that ILT6 directly acts on T-cells.

In consideration of the observation that IFN-gamma is detectable at increased levels during acute phases of MS, it can be deduced from the direct activation of the production of ILT6 by IFN-gamma that ILT6 participates physiologically in the course of inflammatory diseases like autoimmune diseases, e.g. multiple sclerosis. However, the increased production of ILT6 during inflammatory states does not impair the medical application of ILT6 according to the invention.

Claims

1. Human ILT6 protein selected from the natural translation product of the human ILT6 gene and functional derivatives thereof, for medical use.

2. ILT6 according to claim 1, wherein the medical use is directed against diseases selected from autoimmune diseases and undesired immune responses.

3. ILT6 according to claim 2, characterized in that the autoimmune disease is multiple sclerosis, Sjögren's syndrome or an autoimmune disease having acute phases.

4. ILT6 according to claim 2, characterized in that the undesired immune response is the graft versus host disease or the host versus graft disease in transplant patients.

5. ILT6 according to one of the preceding claims, characterized in that the medical use is the reduction of an immune response.

6. ILT6 according to one of claims 2 to 5, characterized in that the immune response is the cellular immune response.

7. ILT6 according to one of claims 2 to 5, characterized in that the immune response is the humoral immune response.

8. ILT6 according to one of the preceding claims, characterized in that ILT6 is the natural translation product from the human ILT6 gene sequence or a functional derivative thereof expressed in eucaryotic or procaryotic cells.

9. Pharmaceutical composition, characterized by comprising ILT6 according to one of the preceding claims.

10. Method for analyzing an immune response or the state of activation of the immune system of a human, characterized in that the method comprises the determination of the concentration of ILT6 in a sample of body fluid obtained from the human.

11. Method according to claim 10, characterized in that the determination of ILT6 is an antibody based assay.

12. Method according to claim 10 or 11, characterized in that the human suffers from an autoimmune disease.

13. Method according to claim 12, characterized in that the autoimmune disease is selected from the group comprising multiple sclerosis and Sjögren's syndrome and autoimmune diseases having acute phases.

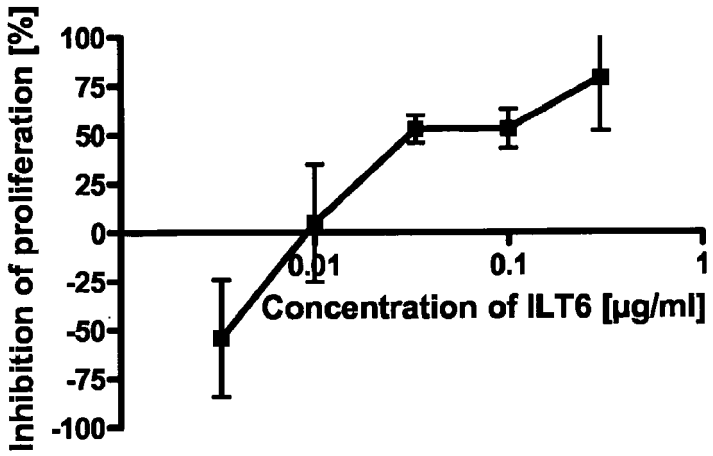
14. Antibody specific for ILT6, characterized in being suitable in a method according to claim 9 to 13.

15. Method for the determination of antibody cross-reactive with ILT6 in a sample of body fluid.

16. ILT6 or a derivative thereof having immunological reactivity derivable from ILT6, for use in a method according to claim 15.

17. Method for analyzing an immune response or the state of activation of the immune system of a human, characterized in that the method comprises the determination of the *in vitro* activity of ILT6 contained in a sample of body fluid obtained from the human in a mixed lymphocyte reaction.

Figure 1:


Inhibition of proliferation [%]
100
75
50
25
0
-25
-50
-75
-100
0.01
0.1
1
Concentration of ILT6 [µg/ml]

SEQUENCE LISTING

<110> Medizinische Hochschule Hannover

Witte, Torsten

Matthias, Torsten

<120> immunomodulation

<130> N1009PCT

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<170> PatentIn version 3.1

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PCT

INTERNATIONAL SEARCH REPORT

(PCT Article 18 and Rules 43 and 44)

Applicant's or agent's file reference	**FOR FURTHER ACTION**	see Form PCT/ISA/220 as well as, where applicable, item 5 below.
N1009PCT		
International application No.	International filing date *(day/month/year)*	(Earliest) Priority Date *(day/month/year)*
PCT/EP2004/052195	15/09/2004	
Applicant		
MEDIZINISCHE HOCHSCHULE HANNOVER		

This International Search Report has been prepared by this International Searching Authority and is transmitted to the applicant according to Article 18. A copy is being transmitted to the International Bureau.

This International Search Report consists of a total of 5 sheets.

[X] It is also accompanied by a copy of each prior art document cited in this report.

1. **Basis of the report**
   a. With regard to the **language,** the international search was carried out on the basis of the international application in the language in which it was filed, unless otherwise indicated under this item.
      [ ] The international search was carried out on the basis of a translation of the international application furnished to this Authority (Rule 23.1(b)).
   b. [X] With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, see Box No. I.

2. [X] **Certain claims were found unsearchable** (See Box II).

3. [ ] **Unity of invention is lacking** (see Box III).

4. With regard to the **title,**
   [ ] the text is approved as submitted by the applicant.
   [X] the text has been established by this Authority to read as follows:

   USE OF ILT6 FOR IMMUNOMODULATION

5. With regard to the **abstract,**
   [X] the text is approved as submitted by the applicant.
   [ ] the text has been established, according to Rule 38.2(b), by this Authority as it appears in Box No. IV. The applicant may, within one month from the date of mailing of this international search report, submit comments to this Authority.

6. With regard to the **drawings,**
   a. the figure of the **drawings** to be published with the abstract is Figure No. _____
      [ ] as suggested by the applicant.
      [ ] as selected by this Authority, because the applicant failed to suggest a figure.
      [ ] as selected by this Authority, because this figure better characterizes the invention.
   b. [X] none of the figures is to be published with the abstract.

INTERNATIONAL SEARCH REPORT

International Application No
PCT/EP2004/052195

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C07K16/28 A61K39/00 G01N33/53

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 7 C07K A61K G01N

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)

EPO-Internal, WPI Data, PAJ, BIOSIS, EMBASE, MEDLINE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WISNIEWSKI ANDRZEJ ET AL: "Distribution of LILRA3 (ILT6/LIR4) deletion in psoriatic patients and healthy controls." HUMAN IMMUNOLOGY, vol. 64, no. 4, April 2003 (2003-04), pages 458-461, XP002326563 ISSN: 0198-8859 *Whole document, in particular: Abstract; page 458, right hand column, last paragraph*	1-17

-/--

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

[ ] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"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)

"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

"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

"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

"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.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
28 April 2005	10/06/2005

Name and mailing address of the ISA	Authorized officer
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	BULCAO DE MELO BARRE

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP2004/052195

Box II Observations where certain claims were found unsearchable (Continuation of item 2 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.: 10-13, 15 and 17
because they relate to subject matter not required to be searched by this Authority, namely:

Although claims 10-13, 15 and 17 are directed to a diagnostic method practised on 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 III Observations where unity of invention is lacking (Continuation of item 3 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 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.:

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/EP2004/052195

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	MOODIE S J ET AL: "Analysis of candidate genes on chromosome 19 in coeliac disease: An association study of the KIR and LILR gene clusters" EUROPEAN JOURNAL OF IMMUNOGENETICS, vol. 29, no. 4, August 2002 (2002-08), pages 287-291, XP002326564 ISSN: 0960-7420	1-9,16
Y	*Whole document, in particular: Abstract*	10-15,17
X	TORKAR MICHAELA ET AL: "Arrangement of the ILT gene cluster: A common null allele of the ILT6 gene results from a 6.7-kbp deletion" EUROPEAN JOURNAL OF IMMUNOLOGY, vol. 30, no. 12, December 2000 (2000-12), pages 3655-3662, XP002326565 ISSN: 0014-2980 cited in the application	1-9,16
Y	*Whole document, in particular: Abstract; page 3657, left hand column, second paragraph and right hand column*	10-15,17
Y	PERICAK-VANCE M A ET AL: "Linkage and association analysis of chromosome 19q13 in multiple sclerosis." NEUROGENETICS. OCT 2001, vol. 3, no. 4, October 2001 (2001-10), pages 195-201, XP002326566 ISSN: 1364-6745 *Whole document, in particular: Abstract; page 199, right hand column, second paragraph*	10-15,17

专利名称(译)	使用ilt6进行免疫调节		
公开(公告)号	EP1789450A1	公开(公告)日	2007-05-30
申请号	EP2004787148	申请日	2004-09-15
[标]申请(专利权)人(译)	MATTHIAS TORSTEN		
申请(专利权)人(译)	MEDIZINISCHE Hochschule的HANNOVER MATTHIAS，TORSTEN		
当前申请(专利权)人(译)	MEDIZINISCHE Hochschule的HANNOVER MATTHIAS，TORSTEN		
[标]发明人	WITTE TORSTEN MATTHIAS TORSTEN		
发明人	WITTE, TORSTEN MATTHIAS, TORSTEN		
IPC分类号	C07K16/28 A61K39/00 G01N33/53		
CPC分类号	A61P1/02 A61P25/00 C07K14/70503 C07K2319/21		
外部链接	Espacenet		

摘要(译)

本发明涉及人类免疫应答的调节。更详细地，本发明涉及ILT6用于调节免疫应答的医学用途，以及含有ILT6的药物组合物。在另一方面，本发明涉及使用ILT6调节其免疫应答的人的医学治疗，例如针对异源和/或自体抗原。此外，本发明涉及用于诊断目的的分析ILT6和可用于该分析的诊断组合物。