[NPL 16] Tomita Y *et al.*, Cancer Sci. 2011 Jan; 102(1): 71-8

Summary of the Invention

The present invention relates to peptides that can induce CTLs specific to KOC1-expressing cells. When these peptides are presented on antigen-presenting cells (APCs) by the human leukocyte antigen (HLA), CTLs that show a specific cytotoxic activity against KOC1-expressing cells are induced. KOC1-derived peptides that have been identified so far to have CTL-inducing ability (CTL inducibility) are either HLA-A2-restricted or HLA-A24-restricted peptides, and cannot induce CTLs against cells that do not express these HLAs. Therefore, conventional peptides are not suitable for performing immunotherapy in subjects that do not have these HLAs. HLA-A11 and HLA-A33 are alleles commonly seen in Asians (Sette A, Sidney J., Immunogenetics 1999, 50: 201-12), and HLA-A03 and HLA-A01 are alleles commonly seen in Caucasians (Cao *et al.*, Hum Immunol 2001; 62(9): 1009-30). It is desirable to administer HLA-A11-restricted peptides to HLA-A11-positive subjects, HLA-A33-restricted peptides to HLA-A33-positive subjects, HLA-A03-restricted peptides to HLA-A03-positive subjects, and HLA-A01-restricted peptides to HLA-A01-positive subjects. Hence, the present invention relates to KOC1-derived peptides with CTL-inducing ability that are restrictive to HLA-A11, HLA-A33, HLA-A03, or HLA-A01. Based on results disclosed herein, the peptides of the present invention have been proven to be epitope peptides that can induce a potent and specific immune response against cells expressing KOC1 and HLA-A11, HLA-A33, HLA-A03, or HLA-A01.

Therefore, one of the objectives of the present invention is to provide KOC1-derived peptides that can induce CTLs in an HLA-A11-, HLA-A33-, HLA-A03- or HLA-A01-restrictive manner. These peptides can be used to induce CTLs *in vitro*, *ex vivo* or *in vivo*, or can be used to administer to subjects for the purpose of inducing an immune response against KOC1-expressing cancer cells. Preferable peptides are peptides comprising the amino acid sequence selected from among SEQ ID NOs: 5, 28, 30, 32, 61, 62, 63, 64, 67, 74,77, 52, 79, 80, 85, 27, 86, 87, 90, 92, 46, 95 and 41; more preferable peptides are nonapeptides or decapeptides; and even more preferable peptides are peptides consisting of the amino acid sequence selected from among SEQ ID NOs: 5, 28, 30, 32, 61, 62, 63, 64, 67, 74,77, 52, 79, 80, 85, 27, 86, 87, 90, 92, 46, 95 and 41.

The peptides of the present invention encompass peptides in which one, two or more amino acid(s) is/are substituted, deleted, inserted and/or added, as long as the resultant modified peptides retain the CTL-inducing ability of the original peptide.

The present invention further provides isolated polynucleotides encoding any one of the peptides of the present invention. Similar to the peptides of the present invention, these

pharmaceutical compositions. In addition, CTLs of the present invention targeting any one of the peptides of the present invention can also be used as an active ingredient of the pharmaceutical compositions of the present invention. The pharmaceutical compositions of the present invention may comprise a therapeutically effective amount or a pharmaceutically effective amount of the above-described active ingredient.

The pharmaceutical compositions of the present invention may also be used as a vaccine. In the context of the present invention, the term "vaccine" (also called "immunogenic composition") refers to a composition that has a function of inducing an immune response that leads to antitumor action when inoculated into an animal. Thus, a pharmaceutical composition of the present invention can be used to induce an immune response that leads to antitumor action. The immune response induced by a peptide, a polynucleotide, an APC, a CTL and a pharmaceutical composition of the present invention is not particularly limited as long as it is an immune response that leads to antitumor action, and examples include induction of cancer cell-specific CTLs and induction of cancer cell-specific cytotoxic activity.

The pharmaceutical compositions of the present invention can be used to treat and/or prevent cancer, and/or prevent postoperative recurrence thereof in human subjects or patients. The pharmaceutical compositions of the present invention can be used preferably to a subject positive for at least one HLA selected from among HLA-A11, HLA-A33, HLA-A03 and HLA-A01. Further, the pharmaceutical compositions of the present invention can be used preferably to treat and/or prevent cancers expressing KOC1 and at least one HLA selected from among HLA-A11, HLA-A33, HLA-A03 and HLA-A01, and/or prevent postoperative recurrence thereof.

In another embodiment, the present invention provides use of an active ingredient selected from below in the manufacture of a pharmaceutical composition for treating or preventing cancer:

(a) a peptide of the present invention;
(b) a polynucleotide encoding a peptide of the present invention in an expressible form;
(c) an APC that presents a peptide of the present invention on its surface;
(d) an exosome that presents a peptide of the present invention on its surface; and
(e) a CTL of the present invention.

Alternatively, the present invention further provides an active ingredient selected from below for use in treating or preventing cancer:

(a) a peptide of the present invention;
(b) a polynucleotide encoding a peptide of the present invention in an expressible form;
(c) an APC that presents a peptide of the present invention on its surface;
(d) an exosome that presents a peptide of the present invention on its surface; and
(e) a CTL of the present invention.

(a) a peptide of the present invention;
(b) a polynucleotide encoding a peptide of the present invention in an expressible form;
(c) an APC that presents on its surface a peptide of the present invention;
(d) an exosome that presents on its surface a peptide of the present invention; and
(e) a CTL of the present invention.

The present invention further provides methods or processes for manufacturing pharmaceutical compositions that induce an immune response against KOC1-expressing cancers, which is a method that may comprise a step of mixing or formulating a peptide or polynucleotide of the present invention with a pharmaceutically acceptable carrier.

Alternatively, the present invention provides methods for inhibiting proliferation of KOC1-expressing cancer cells or methods of inducing an immune response against KOC1-expressing cancers, which comprises a step of administering to a subject vaccines or pharmaceutical compositions comprising an active ingredient selected from below:

(a) a peptide of the present invention;
(b) a polynucleotide encoding a peptide of the present invention in an expressible form;
(c) an APC that presents a peptide of the present invention on its surface;
(d) an exosome that presents a peptide of the present invention on its surface; and
(e) a CTL of the present invention.

In the context of the present invention, KOC1-expressing cancers can be treated by administering a peptide, a polynucleotide, an APC, an exosome and/or a CTL of the present invention. Alternatively, an immune response against KOC1-expressing cancers can be induced by administering a peptide, a polynucleotide, an APC, an exosome and/or a CTL of the present invention. Examples of such cancers include bladder cancer, cervical cancer, cholangiocellular cancer, chronic myeloid leukemia (CML), colon cancer, rectum cancer, esophagus cancer, diffuse gastric cancer, non-small-cell lung cancer, small-cell lung cancer, lymphoma, osteosarcoma, ovarian cancer, kidney cancer, head and neck cancer, soft tissue tumor, testis cancer and such, but are not limited thereto. Further, an immune response against KOC1-expressing cancer cells can be induced by administering a peptide, a polynucleotide, an APC, an exosome and/or a CTL of the present invention. Therefore, before administering a vaccine or pharmaceutical composition comprising an active ingredient described above, it is preferable to confirm whether the level of KOC1 expression at a diseased site in the subject to be treated is augmented or not.

Thus, in one embodiment, the present invention provides a method of treating a KOC1-expressing cancer in a patient in need of the cancer treatment, wherein the method comprises the steps below: