cancer, pancreatic cancer, prostate cancer, renal carcinoma, SCLC and soft tissue tumor. TTK has been identified in bladder cancer, breast cancer, cervical cancer, cholangincellular carcinoma, CML, colorectal cancer, esophageal cancer, liver cancer, NSCLC, lymphoma, osteosarcoma, prostate cancer, SCLC and soft tissue tumor. URLC10 has been identified in bladder cancer, cervical cancer, cholangincellular carcinoma, esophageal cancer, gastric cancer, NSCLC, osteosarcoma, pancreatic cancer and SCLC.

[0009] Summary

The present disclosure is based in part on the discovery of the applicable targets of immunotherapy. Because TAAs have often no immunogenicity, the discovery of appropriate targets is of extreme importance. As noted above, CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK and URLC10 have been identified as up-regulated in various cancers. More particularly, these genes were identified using gene expression profiling with a genome-wide cDNA microarray. As discussed above, expression of CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK and URLC10 has been shown to be specifically up-regulated in various tumor cells, from pancreatic cancer cells to renal cell carcinomas. As described in Table 1, CDH3 expression is validly elevated in 26 out of 34 bladder cancer, 17 out of 19 cervical cancer, all of 19 cholangincellular carcinoma, 30 out of 34 colorectal cancer, 20 out of 21 endometriosis, 13 out of 20 gastric cancer, 7 out of 8 diffuse-type gastric cancer, 36 out of 37 NSCLC, all of 16 pancreatic cancer, all of 21 soft tissue tumor and all of 10 testicular tumor.

[0010] Table 1 further demonstrates that:

EPHA4 expression isvalidly elevated in 14 out of 34 bladder cancer, 8 out of 14 cervical cancer, 10 out of 25 cholangincellular carcinoma, 5 out of 15 endometriosis, 5 out of 8 diffuse-type gastric cancer, all of 5 ovarian cancer, all 14 pancreatic cancer, 20 out of 51 prostate cancer and 14 out of 23 soft tissue tumor.

ECT2 expression is validly elevated in 17 out of 19 bladder cancer, 5 out of 12 breast cancer, all of 14 cervical cancer, all of 13 cholangiocellular carcinoma, all of 5 CML, 7 out of 8 colorectal cancer, 12 out of 16 esophageal cancer, 6 out of 16 NSCLC, 8 out of 10 lymphoma, 1 out of 1 pancreatic cancer, 10 out of 13 prostate cancer, 3 out of 6 renal carcinoma and 12 out of 13 SCLC cancer .

HIG2 expression is validly elevated in 19 out of 20 renal cancer and 7 out of 9 soft tissue tumor.

INHBB expression is validly elevated in 10 out of 21 cholangiocellular carcinoma, all of 12 esophageal cancer, 10 out of 13 NSCLC, 22 out of 24 renal carcinoma, 8 out of 14 SCLC cancer and 45 out of 49 soft tissue tumor .

KIF20A expression is validly elevated in all of 31 bladder cancer, 38 out of 61 breast

cancer, 10 out of 11 cholangiocellular carcinoma, 7 out of 19 esophageal cancer, 21 out of 22 NSCLC, all of 6 ovarian cancer, 17 out of 36 prostate cancer, 6 out of 11 renal carcinoma and all of 15 SCLC .

KNTC2 expression is validly elevated in 30 out of 32 bladder cancer, 47 out of 56 breast cancer, all of 10 cervical cancer, 16 out of 22 cholangioncellular carcinoma, 17 out of 37 CML, 3 out of 10 colorectal cancer, 11 out of 46 esophagus cancer, 15 out of 19 NSCLC, 7 out of 8 lymphoma, 20 out of 24 osteosarcoma, 3 out of 5 ovarian cancer, all of 2 pancreatic cancer, 15 out of 37 prostate cancer, 14 out of 19 renal carcinoma, all of 15 SCLC and 40 out of 59 soft tissue tumor.

TTK expression is validly elevated in all of 27 bladder cancer, 25 out of 30 breast cancer, 15 out of 16 cervical cancer, all of 10 cholangiocellular carcinoma, 5 out of 7 CML, 6 out of 10 colorectal cancer, 24 out of 44 esophageal cancer, 8 out of 15 liver cancer, all of 12 NSCLC, all of 6 lymphoma, 13 out of 16 osteoblastoma, 12 out of 17 prostate cancer, all of 15 SCLC and 16 out of 33 soft tissue tumor.

URLC10 expression is validly elevated in all of 29 bladder cancer, 15 out of 16 cervical cancer, all of 7 cholangiocellular carcinoma, 7 out of 19 esophageal cancer, all of 3 gastric cancer, 24 out of 27 NSCLC, 15 out of 19 osteosarcoma, 4 out of 5 pancreatic cancer, 33 out of 43 soft tissue tumor .

[0011] The present disclosure is based, at least in part, on the identification of specific epitope peptides of the gene products of these genes (CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK and URLC10) which possess the ability to induce cytotoxic T lymphocytes (CTLs) specific to the corresponding molecules. As discussed in detail below, Peripheral Blood Mononuclear Cells (PBMC) of healthy donor were stimulated using HLA-A*2402 or HLA-A*0201 binding candidate peptides derived from CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK or URLC10. CTL clones and/or lines were then established with specific cytotoxicity against the HLA-A24 or HLA-A2 positive target cells pulsed with each of the candidate peptides. These results demonstrate that these peptides are HLA-A24 or HLA-A2 restricted epitope peptides that can induce potent and specific immune responses against cells expressing CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK or URLC10.

[0011a] In a first aspect, there is provided an isolated peptide of less than about 15 amino acids selected from the group consisting of peptides comprising the amino acid sequences of SEQ ID NOs: 288, 271 and 272.

[0011b] In a second aspect, there is provided a peptide having cytotoxic T cell inducibility, wherein said peptide comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 288, 271 and 272, in which 1, 2, or several amino acids are substituted, deleted, or added.[0011c] In a third aspect, there is provided an exosome that presents on its surface a complex comprising any one of the peptides of the first aspect and an HLA antigen.

[0011d] In a fourth aspect, there is provided an antigen-presenting cell, which comprises a complex formed between an HLA antigen and any one of the peptides of the first aspect.

[0012] Accordingly, the present disclosure provides methods for treating or preventing a disease associated with the over-expression of CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK or URLC10, e.g. cancer. Such methods involve the step of administering to a subject in need thereof a CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK and/or URLC10 polypeptides of the disclosure. Administration of such peptide(s) results in the induction of anti-tumor immunity is induced by the administration of these polypeptides. Thus, the present disclosure provides methods for inducing anti-tumor immunity in a subject, such methods involving the step of admin-

istering to the subject the CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK and/or URLC10 polypeptides, as well as pharmaceutical compositions for treating or preventing a disease associated with the over-expression of CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK and/or URLC10, e.g. cancer, that include the CDH3, EPHA4, ECT2, HIG2, INHBB, KIF20A, KNTC2, TTK and URLC10 polypeptides. Examples of such cancers include, but are not limited to., bladder cancer, breast cancer, cervical cancer, cholangincellular carcinoma, CML, colorectal cancer, endometriosis, esophageal cancer, gastric cancer, diffused type gastric cancer, liver cancer, NSCLC, lymphoma, osteosarcoma, ovarian cancer, pancreatic cancer, prostate cancer, renal carcinoma, SCLC, soft tissue tumor and testicular tumor.

[0012a] In a fifth aspect, there is provided a pharmaceutical composition for use in treating or preventing a disease associated with over-expression of the gene of SEQ ID NO: 17, said composition comprising one or more peptides of the first aspect or a polynucleotide encoding the peptides.

[0012b] In a sixth aspect, there is provided an *in vitro* method of inducing an antigen-presenting cell having a high cytotoxic T cell inducibility, said method comprising the step of (a) or (b) below:

(a) contacting an antigen-presenting cell with any one of the peptides of the first aspect; or

(b) transferring a gene comprising a polynucleotide encoding any one of the peptides of the first aspect to an antigen-presenting cell.

[0012c] In a seventh aspect, there is provided an an *in vitro* method of inducing a cytotoxic T cell by contacting a T cell with any one of the peptides of the first aspect.

[0012d] In an eighth aspect, there is provided an *in vitro* method of inducing a cytotoxic T cell, comprising the steps of (a) and (b) below:

(a) contacting an antigen-presenting cell with any one of the peptides of the first aspect; and

(b) mixing the antigen-presenting cell of step (a) with a $CD8^+$ T cell and co-culturing.

[0012e] In a ninth aspect, there is provided an isolated cytotoxic T cell, which is induced by the method of the seventh or eighth aspect, or which is transduced with the nucleic acids encoding the TCR subunits polypeptides binding with any one of the peptides of the first aspect in the context of HLA-A2.

[0012f] In a tenth aspect, there is provided an antigen-presenting cell, which is induced by the method of the sixth aspect.

[0012g] In an eleventh aspect, there is provided a vaccine for use in inhibiting proliferation of cells expressing gene of SEQ ID NO: 17, wherein the vaccine comprises any one of the peptides of the first aspect as the active ingredient.

[0012h] In a twelfth aspect, there is provided use of at least one ingredient selected from the group consisting of (a) to (e) below:
(a) one or more types of any one of the peptides of the first aspect;
(b) one or more types of polynucleotides encoding any one of the peptide(s) of the first aspect in an expressible form;
(c) an APC that presents on its cell surface a complex of any one of the peptides of the first aspect and an HLA antigen;
(d) an exosome that presents on its cell surface a complex of any one of the peptides of the first aspect and an HLA antigen; and
(e) a CTL that targets any one of the peptides of the first aspect
in the manufacture of a medicament for treating or preventing a disease associated with over-expression of the gene of SEQ ID NO: 17.
[0012i] In a thirteenth aspect, there is provided use of any one of the peptides of the first aspect in the manufacture of a vaccine for inhibiting proliferation of cells expressing gene of SEQ ID NO: 17.
[0013]
The present disclosure further provides methods for preventing post-surgery recurrence of the disease mentioned above.
Regarding the specific aims and objectives recited above, it will be understood by those skilled in the art that one or more aspects of this disclosure can meet certain objectives, while one or more other aspects can meet certain other objectives. Each objective may not apply equally, in all its respects, to every aspect of this disclosure. As such, the objects herein can be viewed in the alternative with respect to any one aspect of this disclosure.
[0014]
Additional objects and features of the disclosure will become more fully apparent when the following detailed description is read in conjunction with the accompanying figures and examples. However, it is to be understood that both the foregoing summary of the disclosure and the following detailed description are of preferred embodiments, and not restrictive of the disclosure or other alternate embodiments of the disclosure. In particular, while the disclosure is described herein with reference to a number of specific embodiments, it will be appreciated that the description is illustrative of the disclosure and is not constructed as limiting of the disclosure. Various modifications and applications may occur to those who are skilled in the art, without departing from the spirit and the scope of the disclosure, as described by the appended claims. Likewise, other objects, features, benefits and advantages of the present disclosure will be apparent from this summary and certain embodiments described below, and will be readily apparent to those skilled in the art. Such objects, features, benefits and advantages will be apparent from the above in conjunction with the accompanying examples, data, figures and all reasonable inferences to be drawn therefrom, alone or with consideration of the references incorporated herein.

Brief Description of Drawings

[0015]
Various aspects and applications of the present invention will become apparent to the skilled artisan upon consideration of the brief description of the figures and the

CLAIMS

[Claim 1]

An isolated peptide of less than about 15 amino acids selected from the group consisting of peptides comprising the amino acid sequences of SEQ ID NOs: 288, 271 and 272.

[Claim 2]

A peptide having cytotoxic T cell inducibility, wherein said peptide comprises an amino acid sequence selected from the group consisting of SEQ ID NOs: 288, 271 and 272, in which 1, 2, or several amino acids are substituted, deleted, or added.

[Claim 3]

The peptide of claim 2, wherein the second amino acid from the N-terminus is leucine or methionine.

[Claim 4]

The peptide of claim 2 or 3, wherein the C-terminal amino acid is valine or leucine.

[Claim 5]

The peptide of claim 1, which consists of the amino acid sequence selected from the group consisting of SEQ ID NOs: 288, 271 and 272.

[Claim 6]

A pharmaceutical composition for use in treating or preventing a disease associated with over-expression of the gene of SEQ ID NO: 17, said composition comprising one or more peptides of any one of claims 1 to 5 or a polynucleotide encoding the peptides.

[Claim 7]

The pharmaceutical composition of claim 6, wherein the disease associated with over-expression of the gene of SEQ ID NO: 17 includes cancers.

[Claim 8]

A pharmaceutical composition of claim 7, wherein the cancers include bladder cancer, breast cancer, cervical cancer, cholangiocellular carcinoma, CML, colorectal cancer, endometriosis, esophageal cancer, gastric cancer, diffused type gastric cancer, liver, NSCLC, lymphoma, osteosarcoma, ovarian cancer, pancreatic cancer, prostate cancer, renal carcinoma, SCLC, soft tissue tumor and testicular tumor.

[Claim 9]

An exosome that presents on its surface a complex comprising the peptide of any one of claims 1 to 5 and an HLA antigen.

[Claim 10]

The exosome of claim 9, wherein the HLA antigen is HLA-A2.

[Claim 11]

The exosome of claim 10, wherein the HLA-A2 is HLA-A0201.

[Claim 12]
An *in vitro* method of inducing an antigen-presenting cell having a high cytotoxic T cell inducibility, said method comprising the step of (a) or (b) below:
(a) contacting an antigen-presenting cell with the peptide of any one of claims 1 to 5; or
(b) transferring a gene comprising a polynucleotide encoding the peptide of any one of claims 1 to 5 to an antigen-presenting cell.
[Claim 13]
An *in vitro* method of inducing a cytotoxic T cell by contacting a T cell with the peptide of any one of claims 1 to 5.
[Claim 14]
An *in vitro* method of inducing a cytotoxic T cell, comprising the steps of (a) and (b) below:
(a) contacting an antigen-presenting cell with the peptide of any one of claims 1 to 5; and
(b) mixing the antigen-presenting cell of step (a) with a $CD8^+$ T cell and co-culturing.
[Claim 15]
An isolated cytotoxic T cell, which is induced by the method of claim 13 or 14, or which is transduced with the nucleic acids encoding the TCR subunits polypeptides binding with the peptide of any one of claims 1 to 5 in the context of HLA-A2.
[Claim 16]
An antigen-presenting cell, which comprises a complex formed between an HLA antigen and the peptide of any one of claims 1 to 5.
[Claim 17]
An antigen-presenting cell, which is induced by the method of claim 12.
[Claim 18]
A vaccine for use in inhibiting proliferation of cells expressing gene of SEQ ID NO: 17, wherein the vaccine comprises the peptide of any one of claims 1 to 5 as the active ingredient.
[Claim 19]
The vaccine of claim 18, wherein the cells expressing gene of SEQ ID NO: 17 include cancer cells.
[Claim 20]
The vaccine of claim 19, wherein the cancer cells include cells of bladder cancer, breast cancer, cervical cancer, cholangiocellular carcinoma, CML, colorectal cancer, endometriosis, esophageal cancer, gastric cancer, diffused type gastric cancer, liver, NSCLC, lymphoma, osteosarcoma, ovarian cancer, pancreatic cancer, prostate cancer, renal carcinoma, SCLC, soft tissue tumor and testicular tumor.
[Claim 21]
The vaccine of claim 20, which is formulated for administration to a subject whose HLA antigen

is HLA-A2.

[Claim 22]

The vaccine of claim 21, wherein the HLA-A2 is HLA-A0201.

[Claim 23]

Use of at least one ingredient selected from the group consisting of (a) to (e) below:

(a) one or more types of peptides of any one of claims 1 to 5;

(b) one or more types of polynucleotides encoding the peptide(s) of any one of claims 1 to 5 in an expressible form;

(c) an APC that presents on its cell surface a complex of the peptide of any one of claims 1 to 5 and an HLA antigen;

(d) an exosome that presents on its cell surface a complex of the peptide of any one of claims 1 to 5 and an HLA antigen; and

(e) a CTL that targets the peptide of any one of claims 1 to 5

in the manufacture of a medicament for treating or preventing a disease associated with over-expression of the gene of SEQ ID NO: 17.

[Claim 24]

The use of claim 23, wherein the disease associated with over-expression of the gene of SEQ ID NO: 17 includes cancers.

[Claim 25]

The use of claim 24, wherein the cancers include bladder cancer, breast cancer, cervical cancer, cholangiocellular carcinoma, CML, colorectal cancer, endometriosis, esophageal cancer, gastric cancer, diffused type gastric cancer, liver, NSCLC, lymphoma, osteosarcoma, ovarian cancer, pancreatic cancer, prostate cancer, renal carcinoma, SCLC, soft tissue tumor and testicular tumor.

[Claim 26]

Use of the peptide of any one of claims 1 to 5 in the manufacture of a vaccine for inhibiting proliferation of cells expressing gene of SEQ ID NO: 17.

[Claim 27]

The use of claim 26, wherein the cells expressing gene of SEQ ID NO: 17 include cancer cells.

[Claim 28]

The use of claim 27, wherein the cancer cells include cells of bladder cancer, breast cancer, cervical cancer, cholangiocellular carcinoma, CML, colorectal cancer, endometriosis, esophageal cancer, gastric cancer, diffused type gastric cancer, liver, NSCLC, lymphoma, osteosarcoma, ovarian cancer, pancreatic cancer, prostate cancer, renal carcinoma, SCLC, soft tissue tumor and testicular tumor.