and exatecan are conjugated to each other via the linker, and confirmed an excellent antitumor effect exhibited by the conjugate to thereby complete the present ~~invention~~disclosure.

[0014a] In one aspect, the present disclosure refers to a method for producing an antibody-drug conjugate, in which a drug-linker structure represented by the following formula is conjugated to an anti-HER2 antibody via a thioether bond,

-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2-O-CH_2-C(=O)-(NH-DX)

comprising a step of

treating the anti-HER2 antibody in a reducing condition and thereafter

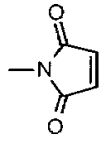
reacting the anti-HER2 antibody with a compound represented by the following formula:

(maleimid-N-yl)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2-O-CH_2-C(=O)-(NH-DX)

wherein,

(maleimid-N-yl)- is a group represented by the following formula:

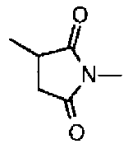
[Formula 1]



wherein the nitrogen atom is a connecting position,

-(Succinimid-3-yl-N)- has a structure represented by the following formula:

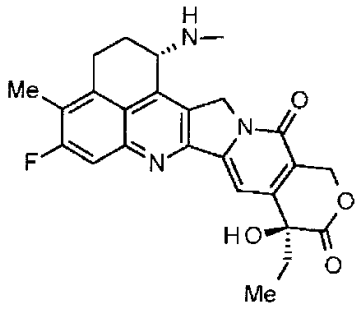
[Formula 2]



which is connected to the antibody at position 3 thereof and is connected to a methylene group in the linker structure containing this structure on the nitrogen atom at position 1,

-(NH-DX) represents a group represented by the following formula:

[Formula 3]



wherein the nitrogen atom of the amino group at position 1 is a connecting position, and

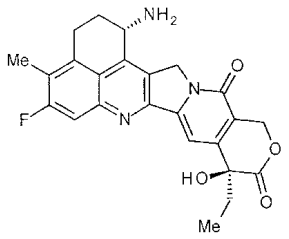
-GGFG- represents a tetrapeptide residue of -Gly-Gly-Phe-Gly-.

[0015]

Specifically, the present ~~invention~~disclosure relates to the following.

[1] An antibody-drug conjugate wherein an antitumor compound represented by the following formula:

[Formula 2]



is conjugated to an anti-HER2 antibody via a linker having a structure represented by the following formula:

$-L^1-L^2-L^P-NH-(CH_2)n^1-L^a-(CH_2)n^2-C(=O)-$

via a thioether bond which is formed at a disulfide bond moiety present in the hinge part of the anti-HER2 antibody.

[0016]

Here, the anti-HER2 antibody is connected to the terminal L^1,

the antitumor compound is connected to the carbonyl group of the $-(CH_2)n^2-C(=O)-$ moiety with the nitrogen

atom of the amino group at position 1 as the connecting position,

wherein

n^1 represents an integer of 0 to 6,

n^2 represents an integer of 0 to 5,

L^1 represents -(Succinimid-3-yl-N)-$(CH_2)n^3$-C(=O)-,

wherein n^3 represents an integer of 2 to 8,

L^2 represents -NH-$(CH_2CH_2-O)n^4$-CH_2CH_2-C(=O)- or a single bond,

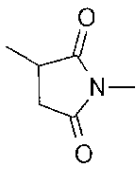
wherein n^4 represents an integer of 1 to 6,

L^P represents a peptide residue consisting of 2 to 7 amino acids,

L^a represents -O- or a single bond, and

-(Succinimid-3-yl-N)- has a structure represented by the following formula:

[Formula 3]



which is connected to the anti-HER2 antibody at position 3 thereof and is connected to the methylene group in the linker structure containing this structure on the nitrogen atom at position 1.

[0017]

The present ~~invention~~disclosure further relates to each of the following.

Claims

[Claim 1]

A method for producing an antibody-drug conjugate, in which a drug-linker structure represented by the following formula is conjugated to an anti-HER2 antibody via a thioether bond,

-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2-O-CH_2-C(=O)-(NH-DX)

comprising a step of

treating the anti-HER2 antibody in a reducing condition and thereafter

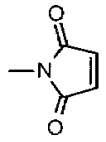
reacting the anti-HER2 antibody with a compound represented by the following formula:

(maleimid-N-yl)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2-O-CH_2-C(=O)-(NH-DX)

wherein,

(maleimid-N-yl)- is a group represented by the following formula:

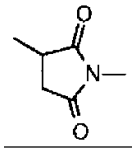
[Formula 1]



wherein the nitrogen atom is a connecting position,

-(Succinimid-3-yl-N)- has a structure represented by the following formula:

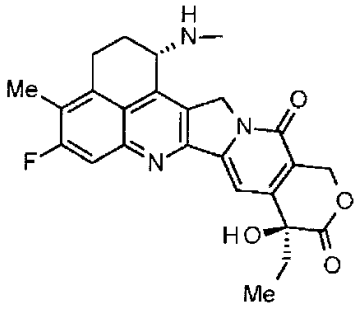
[Formula 2]



which is connected to the antibody at position 3 thereof and is connected to a methylene group in the linker structure containing this structure on the nitrogen atom at position 1,

-(NH-DX) represents a group represented by the following formula:

[Formula 3]



wherein the nitrogen atom of the amino group at position 1 is a connecting position, and

-GGFG- represents a tetrapeptide residue of -Gly-Gly-Phe-Gly-.

[Claim 2]

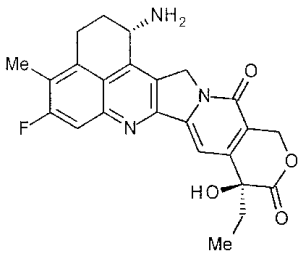
The method according to claim 1, wherein an average number of units of the drug-linker structure conjugated per antibody is in a range of from 2 to 8.

[Claim 3]

The method according to claim 1, wherein an average number of units of the drug-linker structure conjugated per antibody is in a range of from 3 to 8.

~~An antibody-drug conjugate wherein an antitumor compound represented by the following formula:~~

~~[Formula 1]~~



~~is conjugated to an anti-HER2 antibody via a linker having a structure represented by the following formula:~~

~~$-L^1-L^2-L^P-NH-(CH_2)n^1-L^a-(CH_2)n^2-C(=O)-$~~

~~via a thioether bond which is formed at a disulfide bond moiety present in the hinge part of the anti-HER2 antibody,~~

~~wherein~~

~~the anti-HER2 antibody is connected to the terminal L^1,~~

~~the antitumor compound is connected to the carbonyl group of the $-(CH_2)n^2-C(=O)-$ moiety with the nitrogen atom of the amino group at position 1 as the connecting position,~~

~~wherein~~

~~n^1 represents an integer of 0 to 6,~~

~~n^2 represents an integer of 0 to 5,~~

~~L^1 represents $-(Succinimid-3-yl-N)-(CH_2)n^3-C(=O)-$,~~

~~wherein n^3 represents an integer of 2 to 8,~~

~~L^2 represents $-NH-(CH_2CH_2-O)n^4-CH_2CH_2-C(=O)-$ or a single bond,~~

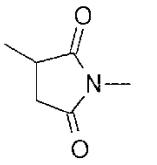
wherein n^4 represents an integer of 1 to 6,

L^P represents a peptide residue consisting of 2 to 7 amino acids,

L^a represents -O- or a single bond, and

-(Succinimid-3-yl-N)- has a structure represented by the following formula:

[Formula 2]



which is connected to the anti-HER2 antibody at position 3 thereof and is connected to the methylene group in the linker structure containing this structure on the nitrogen atom at position 1.

[Claim 2]

The antibody-drug conjugate according to claim 1, wherein the peptide residue L^P is a peptide residue comprising an amino acid selected from phenylalanine, glycine, valine, lysine, citrulline, serine, glutamic acid, and aspartic acid.

[Claim 3]

The antibody-drug conjugate according to claim 1 or 2, wherein L^P is a peptide residue selected from the following group:

-GGF-

-DGGF-

-(D-)D-GGF-

~~-EGGF-~~

~~-GGFG-~~

~~-SGGF-~~

~~-KGGF-~~

~~-DGGFG-~~

~~-GGFGG-~~

~~-DDGGFG-~~

~~-KDGGFG-~~

~~-GGFGGGF-~~

~~wherein "(D-)D" represents D-aspartic acid.~~

~~[Claim 4]~~

~~The antibody-drug conjugate according to claim 1 or 2, wherein L^P is a peptide residue consisting of 4 amino acids.~~

~~[Claim 5]~~

~~The antibody drug conjugate according to any one of claims 1 to 4, wherein L^P is the tetrapeptide residue -GGFG-.~~

~~[Claim 6]~~

~~The antibody-drug conjugate according to any one of claims 1 to 5, wherein n^3 is an integer of 2 to 5, and L^2 is a single bond.~~

~~[Claim 7]~~

~~The antibody drug conjugate according to any one of claims 1 to 5, wherein n^3 is an integer of 2 to 5, L^2 is $-NH-(CH_2CH_2-O)n^4-CH_2CH_2-C(=O)-$, and n^4 is 2 or 4.~~

~~[Claim 8]~~

~~The antibody-drug conjugate according to any one of claims 1 to 7, wherein $-NH-(CH_2)n^1-L^a-(CH_2)n^2-C(=O)-$ is a partial structure having a chain length of 4 to 7 atoms.~~

~~[Claim 9]~~

~~The antibody drug conjugate according to any one of claims 1 to 7, wherein $-NH-(CH_2)n^1-L^a-(CH_2)n^2-C(=O)-$ is a partial structure having a chain length of 5 or 6 atoms.~~

~~[Claim 10]~~

~~The antibody-drug conjugate according to any one of claims 1 to 9, wherein $-NH-(CH_2)n^1-L^a-(CH_2)n^2-C(=O)-$ is any of the following group:~~

~~$-NH-CH_2CH_2-C(=O)-$~~

~~$-NH-CH_2CH_2CH_2-C(=O)-$~~

~~$-NH-CH_2CH_2CH_2CH_2-C(=O)-$~~

~~$-NH-CH_2CH_2CH_2CH_2CH_2-C(=O)-$~~

~~$-NH-CH_2-O-CH_2-C(=O)-$~~

~~$-NH-CH_2CH_2-O-CH_2-C(=O)-$~~

~~$-NH-CH_2CH_2-O-C(=O)-$~~

~~[Claim 11]~~

~~The antibody-drug conjugate according to claim 10, wherein $-NH-(CH_2)n^1-L^a-(CH_2)n^2-C(=O)-$ is any of the following group:~~

~~$-NH-CH_2CH_2CH_2-C(=O)-$~~

~~$-NH-CH_2-O-CH_2-C(=O)-$~~

~~$-NH-CH_2CH_2-O-CH_2-C(=O)-$~~

~~[Claim 12]~~

~~The antibody-drug conjugate according to any one of claims 1 to 9, wherein the drug-linker structure moiety having the drug connected to -L^1-L^2-L^P-NH-$(CH_2)n^1$-L^a-$(CH_2)n^2$-C(=O)- is one drug-linker structure selected from the following group:~~

~~-(Succinimid-3-yl-N)-CH_2CH_2-C(=O)-GGFG-NH-CH_2CH_2-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-CH_2CH_2-C(=O)-GGFG-NH-$CH_2CH_2CH_2$-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2CH_2-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-$CH_2CH_2CH_2$-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2-O-CH_2-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2CH_2-O-CH_2-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2CH_2-O-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-CH_2CH_2-C(=O)-NH-CH_2CH_2-O-CH_2CH_2-O-CH_2CH_2-C(=O)-GGFG-NH-CH_2CH_2-C(=O)-(NH-DX)~~

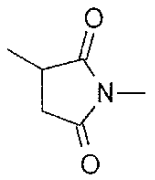
~~-(Succinimid-3-yl-N)-CH_2CH_2-C(=O)-NH-CH_2CH_2-O-CH_2CH_2-O-CH_2CH_2-C(=O)-GGFG-NH-$CH_2CH_2CH_2$-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-CH_2CH_2-C(=O)-NH-CH_2CH_2-O-CH_2CH_2-O-CH_2CH_2-O-CH_2CH_2-O-CH_2CH_2-C(=O)-GGFG-NH-CH_2CH_2-C(=O)-(NH-DX)~~

~~-(Succinimid-3-yl-N)-CH_2CH_2-C(=O)-NH-CH_2CH_2-O-CH_2CH_2-O-CH_2CH_2-O-CH_2CH_2-O-CH_2CH_2-C(=O)-GGFG-NH-$CH_2CH_2CH_2$-C(=O)-(NH-DX)~~

~~wherein -(Succinimid-3-yl-N)- has a structure represented by the following formula:~~

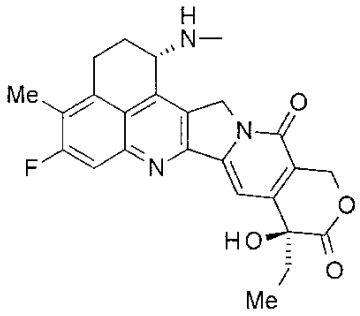
~~[Formula 3]~~



~~which is connected to the anti-HER2 antibody at position 3 thereof and is connected to the methylene group in the linker structure containing this structure on the nitrogen atom at position 1,~~

~~-(NH-DX) represents a group represented by the following formula:~~

~~[Formula 4]~~



~~wherein the nitrogen atom of the amino group at position 1 is the connecting position, and~~

~~-GGFG- represents the tetrapeptide residue -Gly-Gly-Phe-Gly-.~~

[Claim 13]

The antibody-drug conjugate according to claim 12, wherein the drug-linker structure moiety is one drug-linker structure selected from the following group:

-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2-O-CH_2-C(=O)-(NH-DX)

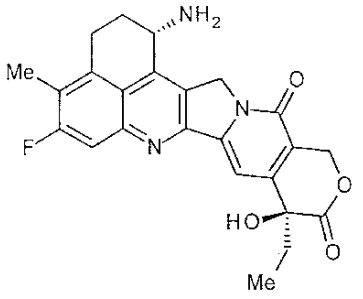
-(Succinimid-3-yl-N)-$CH_2CH_2CH_2CH_2CH_2$-C(=O)-GGFG-NH-CH_2CH_2-O-CH_2-C(=O)-(NH-DX)

-(Succinimid-3-yl-N)-CH_2CH_2-C(=O)-NH-CH_2CH_2-O-CH_2CH_2-O-CH_2CH_2-C(=O)-GGFG-NH-$CH_2CH_2CH_2$-C(=O)-(NH-DX)

[Claim 14]

An antibody-drug conjugate wherein an antitumor compound represented by the following formula:

[Formula 5]



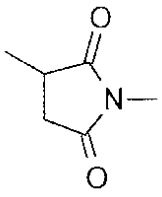
is conjugated to an anti-HER2 antibody via a linker having a structure represented by the following formula:

-L^1-L^2-L^P-NH-$(CH_2)n^1$-L^a-$(CH_2)n^2$-C(=O)-

via a thioether bond which is formed at a disulfide bond moiety present in the hinge part of the anti-HER2 antibody,

wherein

the anti-HER2 antibody is connected to the terminal L^1,
the antitumor compound is connected to the carbonyl group of the $-(CH_2)n^2-C(=O)-$ moiety,
wherein
n^1 represents an integer of 0 to 6,
n^2 represents an integer of 0 to 5,
L^1 represents -(Succinimid-3-yl-N)-$(CH_2)n^3-C(=O)-$,
wherein n^3 represents an integer of 2 to 8,
L^2 represents $-NH-(CH_2CH_2-O)n^4-CH_2CH_2-C(=O)-$ or a single bond,
wherein n^4 represents an integer of 1 to 6,
L^P represents the tetrapeptide residue -GGFG-,
L^a represents -O- or a single bond, and
-(Succinimid-3-yl-N)- has a structure represented by the following formula:
[Formula 6]



which is connected to the anti-HER2 antibody at position 3 thereof and is connected to the methylene group in the linker structure containing this structure on the nitrogen atom at position 1.
[Claim 15]
The antibody-drug conjugate according to claim 14, wherein

~~n^1 is 3, n^2 is 0, n^3 is 2, L^2 is -NH-$(CH_2CH_2-O)n^4$-CH_2CH_2-C(=O)-, n^4 is 2, and L^a is a single bond,~~

~~n^1 is 1, n^2 is 1, n^3 is 5, L^2 is a single bond, and L^a is -O-, or~~

~~n^1 is 2, n^2 is 1, n^3 is 5, L^2 is a single bond, and L^a is -O-.~~

~~[Claim 16]~~

~~The antibody-drug conjugate according to claim 14 or 15, wherein n^3 is 2 or 5, and L^2 is a single bond.~~

~~[Claim 17]~~

~~The antibody-drug conjugate according to claim 14 or 15, wherein n^3 is 2 or 5, L^2 is -NH-$(CH_2CH_2-O)n^4$-CH_2CH_2-C(=O)-, and n^4 is 2 or 4.~~

~~[Claim 18]~~

~~The antibody-drug conjugate according to any one of claims 14 to 17, wherein -NH-$(CH_2)n^1$-L^a-$(CH_2)n^2$-C(=O)- is any of the following group:~~

~~-NH-$CH_2CH_2CH_2$-C(=O)-~~

~~-NH-CH_2-O-CH_2-C(=O)-~~

~~-NH-CH_2CH_2-O-CH_2-C(=O)-~~

~~[Claim 19]~~

~~The antibody-drug conjugate according to any one of claims 1 to 18, wherein the average number of units of the selected one drug linker structure conjugated per antibody molecule is in the range of from 1 to 10.~~

~~[Claim 20]~~

~~The antibody-drug conjugate according to any one of claims 1 to 18, wherein the average number of units~~

~~of the selected one drug-linker structure conjugated per antibody molecule is in the range of from 2 to 8.~~

~~[Claim 21]~~

~~The antibody-drug conjugate according to any one of claims 1 to 18, wherein the average number of units of the selected one drug linker structure conjugated per antibody molecule is in the range of from 3 to 8.~~

~~[Claim 22]~~

~~A drug containing the antibody-drug conjugate according to any one of claims 1 to 21, a salt thereof, or a hydrate thereof.~~

~~[Claim 23]~~

~~An antitumor drug and/or anticancer drug containing the antibody-drug conjugate according to any one of claims 1 to 21, a salt thereof, or a hydrate thereof.~~

~~[Claim 24]~~

~~The antitumor drug and/or anticancer drug according to claim 23, which is for use against lung cancer, urothelial cancer, colorectal cancer, prostate cancer, ovarian cancer, pancreatic cancer, breast cancer, bladder cancer, gastric cancer, gastrointestinal stromal tumor, uterine cervix cancer, esophageal cancer, squamous cell carcinoma, peritoneal cancer, liver cancer, hepatocellular cancer, colon cancer, rectal cancer, colorectal cancer, endometrial cancer, uterine cancer, salivary gland cancer, kidney cancer, vulval cancer, thyroid cancer, penis cancer,~~

~~leukemia, malignant lymphoma, plasmacytoma, myeloma, or sarcoma.~~

~~[Claim 25]~~

~~A pharmaceutical composition containing the antibody-drug conjugate according to any one of claims 1 to 21, a salt thereof, or a hydrate thereof as an active component, and a pharmaceutically acceptable formulation component.~~

~~[Claim 26]~~

~~The pharmaceutical composition according to claim 25, which is for use against lung cancer, urothelial cancer, colorectal cancer, prostate cancer, ovarian cancer, pancreatic cancer, breast cancer, bladder cancer, gastric cancer, gastrointestinal stromal tumor, uterine cervix cancer, esophageal cancer, squamous cell carcinoma, peritoneal cancer, liver cancer, hepatocellular cancer, colon cancer, rectal cancer, colorectal cancer, endometrial cancer, uterine cancer, salivary gland cancer, kidney cancer, vulval cancer, thyroid cancer, penis cancer, leukemia, malignant lymphoma, plasmacytoma, myeloma, or sarcoma.~~

~~[Claim 27]~~

~~A method for treating tumor and/or cancer comprising administering the antibody drug conjugate according to any one of claims 1 to 21, a salt thereof, or a hydrate thereof.~~