Patent Document 2: Japanese Translation of PCT Application No. 2013-539362

Patent Document 3: Japanese Translation of PCT Application No. 2008-514201

Patent Document 4: Japanese Translation of PCT Application No. 2009-504164

Patent Document 5: Japanese Translation of PCT Application No. 2009-541361

Patent Document 6: Japanese Translation of PCT Application No. 2009-540837

Summary

Technical Problem

[0007] An object of the present invention is to provide a method of presenting a cyclic peptide on a protein having a loop structure with the protein as a scaffold while retaining the structure and activity which the cyclic peptide has.

Solution to Problem

[0007a] In one aspect, the present disclosure refers to a method of presenting a cyclic peptide, having a binding activity to an intended molecule, on a protein having a loop structure, comprising: replacing the amino acid residues constituting the loop structure with the cyclic peptide, and wherein the two amino acid residues for forming a chemically crosslinked structure in the cyclic peptide are replaced with two amino acid residues constituting the loop structure; fusing the cyclic peptide to the protein having the loop structure, while retaining the binding activity to the intended molecule; and wherein the two amino acid residues constituting the loop structure have a Cα atomic distance within a range of from 4 to 7Å.

[0007b] In another aspect, the present disclosure refers to a method of fusing a cyclic peptide, having a binding activity to an intended molecule, to a protein having a loop structure, thereby producing a modified protein having the cyclic peptide presented on the protein, comprising: replacing the amino acid residues constituting the loop structure with the cyclic peptide, and wherein the two amino acid residues for forming a chemically crosslinked structure in the cyclic peptide are replaced with two amino acid residues constituting the loop structure; and fusing the cyclic peptide to the protein having the loop structure, while retaining the binding activity to the intended molecule, wherein the fusing step comprises: selecting, from an amino acid sequence of the cyclic peptide, a partial amino acid sequence to be presented on the protein and selecting a base sequence in the protein having a loop structure corresponding to the partial amino acid sequence, selecting base sequences corresponding to two amino acid sequences constituting the loop structure of the protein having a loop structure, respectively, removing a base present between the selected base sequences if necessary, and preparing a nucleic acid having, inserted and incorporated therein, the base sequence corresponding to the partial amino acid sequence of the selected cyclic peptide, and translating the nucleic acid wherein the modified protein retains the binding activity to the intended molecule of the cyclic peptide; and wherein the two amino acid residues constituting the loop structure have a Cα atomic distance within a range of from 4 to 7Å.

[0008] The following are the details of the present disclosure.

(1) A method of presenting a cyclic peptide on a protein having a loop structure, includes:

using, as the cyclic peptide, that having a chemically crosslinked structure for forming an intramolecular cyclic structure, and

replacing the chemically crosslinked structure of the cyclic peptide with two amino acid residues constituting the loop structure and fusing the cyclic peptide to the protein having a loop structure.

What is claimed is:

1. A method of presenting a cyclic peptide, having a binding activity to an intended molecule, on a protein having a loop structure, comprising:

replacing the amino acid residues constituting the loop structure with the cyclic peptide, and

wherein the two amino acid residues for forming a chemically crosslinked structure in the cyclic peptide are replaced with two amino acid residues constituting the loop structure;

fusing the cyclic peptide to the protein having the loop structure, while retaining the binding activity to the intended molecule; and

wherein the two amino acid residues constituting the loop structure have a Cα atomic distance within a range of from 4 to 7Å.

2. The method according to Claim 1, wherein the protein having a loop structure has from 1 to 15 amino acid residues between the two amino acid residues constituting the loop structure.

3. The method according to Claim 1 or 2, wherein the cyclic peptide is composed of proteinogenic amino acids and/or non-proteinogenic amino acids.

4. The method according to any one of Claims 1 to 3, wherein the chemically crosslinked structure contains a thioether bond or a disulfide bond.

5. The method according to any one of Claims 1 to 4, wherein two or more cyclic peptides each fuse to respectively different loop structures of a protein having a plurality of loop structures.

6. The method according to Claim 5, wherein the two or more cyclic peptides have the same amino acid sequence.

7. The method according to Claim 5, wherein the two or more cyclic peptides have respectively different amino acid sequences.

8. The method according to any one of Claims 1 to 7, wherein in replacing the chemically crosslinked structure of the cyclic peptide with the two amino acid residues constituting the loop structure, the chemically crosslinked structure of the cyclic peptide is replaced, via a linker sequence, with the two amino acid residues constituting the loop structure.

9. The method according to any one of Claims 1 to 8, wherein the chemically crosslinked structure of the cyclic peptide is replaced with the two amino acid residues constituting the loop structure while including an amino acid sequence constituting the loop structure other than the two amino acids constituting the loop structure.

10. A method of fusing a cyclic peptide, having a binding activity to an intended molecule, to a protein having a loop structure, thereby producing a modified protein having the cyclic peptide presented on the protein, comprising:

replacing the amino acid residues constituting the loop structure with the cyclic peptide, and

wherein the two amino acid residues for forming a chemically crosslinked structure in the cyclic peptide are replaced with two amino acid residues constituting the loop structure; and

fusing the cyclic peptide to the protein having the loop structure, while retaining the binding activity to the intended molecule,

wherein the fusing step comprises:

selecting, from an amino acid sequence of the cyclic peptide, a partial amino acid sequence to be presented on the protein and selecting a base sequence in the protein having a loop structure corresponding to the partial amino acid sequence,

selecting base sequences corresponding to two amino acid sequences constituting the loop structure of the protein having a loop structure, respectively, removing a base present between the selected base sequences if necessary, and preparing a nucleic acid having, inserted and incorporated therein, the base sequence corresponding to the partial amino acid sequence of the selected cyclic peptide, and

translating the nucleic acid

wherein the modified protein retains the binding activity to the intended molecule of the cyclic peptide; and

wherein the two amino acid residues constituting the loop structure have a Cα atomic distance within a range of from 4 to 7Å.