CLAIMS

1. A modified biotin-binding protein comprising an amino acid sequence represented by SEQ ID NO: 2, or an amino acid sequence having an identity of not less than 95% to the amino acid sequence of SEQ ID NO: 2 and having biotin-binding activity, provided that the modified biotin-binding has an asparagine residue at position 115 of SEQ ID NO: 2 replaced with cysteine, wherein the modified biotin-binding protein maintains biotin-binding activity of not less than 75% after a heat treatment at 99.9°C for 30 min, compared with that of before the treatment.

2. The modified biotin-binding protein according to Claim 1, wherein the modified biotin-binding protein maintains biotin-binding activity of not less than 50% after a treatment in 60% aprotic polar organic solvent for 30 min, compared with that of before the treatment.

3. The modified biotin-binding protein according to Claim 2, wherein the aprotic polar organic solvent is dimethyl sulfoxide.

4. The modified biotin-binding protein according to any one of Claims 1 to 3, wherein the modified biotin-binding protein comprises an amino acid having an identity of not less than 98% to the amino acid sequence of SEQ ID NO: 2.

5. A modified biotin-binding protein comprising an amino acid sequence represented by SEQ ID NO: 2, provided that the modified biotin-binding protein has an asparagine residue at position 115 of SEQ ID NO: 2 replaced with cysteine (TM2-N115C).

6. A support to which the protein according to any one of Claims 1 to 5 is immobilized.

7. A nucleic acid encoding the protein according to any one of Claims 1 to 5.

8. A vector containing the nucleic acid according to Claim 7.

9. A method for separation, concentration, capture, purification, and/or detection of the protein according to any one of Claims 1 to 5, which comprises the following steps:

1) heat-treating a sample containing the protein at a temperature of at least 90°C for

~~at least 10 min; and~~

~~2) collecting the protein which does not receive heat-denature in step 1) to thereby separate, concentrate, capture, or purify the protein, and/or detect the protein.~~

CLAIMS

~~10.~~1. A method for separation, concentration, capture, purification and/or detection of a biotin-linked substance, which comprises the following steps:

1) contacting ~~the~~ a support, ~~according to Claim 6~~to which a modified biotin-binding protein is immobilized, with a biotin-linked substance, whereby the biotin-linked substance is bound to the support;

2) washing off contaminants which are not bound to the support with a solution containing 60% to 80% of aprotic polar organic solvent; and

3) collecting the biotin-linked substance which is bound to the support to thereby separate, concentrate, capture or purify the substance and/or detect the substance~~.~~; wherein the modified biotin-binding protein comprises an amino acid sequence represented by SEQ ID NO: 2 or an amino acid sequence having an identity of not less than 95% to the amino acid sequence of SEQ ID NO: 2, and has biotin-binding activity, provided that the modified biotin-binding protein has an asparagine residue at position 115 of SEQ ID NO: 2 replaced with cysteine, wherein the modified biotin-binding protein maintains biotin-binding activity of not less than 75% after a heat treatment at 99.9°C for 30 min, compared with that of before the treatment.

~~11.~~2. The method according to Claim ~~10~~1, wherein the aprotic polar organic solvent is dimethyl sulfoxide.

3. The method according to Claim 1 or 2, wherein the modified biotin-binding protein maintains biotin-binding activity of not less than 50% after a treatment in 60% aprotic polar organic solvent for 30 min, compared with that of before the treatment.

4. The method according to any one of Claims 1 to 3, wherein the modified biotin-binding protein comprises an amino acid having an identity of not less than 98% to the amino acid sequence of SEQ ID NO: 2.