

Non Patent Literature

- [0030] [NPL1] Reichert et al., Nat. Biotechnol. 23:1073-1078 (2005)
- [NPL2] Pavlou et al., Eur. J. Pharm. Biopharm. 59(3):389-396 (2005)
- [NPL3] Kim et al., Mol. Cells. 20 (1):17-29 (2005)
- [NPL4] Hinton et al., J. Immunol. 176 (1):346-356 (2006)
- [NPL5] Ghetie et al., Nat. Biotechnol. 15(7):637-640 (1997))
- [NPL6] Rajpal et al., Proc. Natl. Acad. Sci. USA 102(24):8466-8471 (2005)
- [NPL7] Wu et al., J. Mol. Biol. 368:652 (2007)
- [NPL8] Wu et al., J. Mol. Biol. 368:652-665 (2007)
- [NPL9] Igawa et al., Nat. Biotechnol. 28:1203-1207 (2010)
- [NPL10] Dall'Acqua et al., J. Biol. Chem. 281:23514-235249 (2006)
- [NPL11] Zalevsky et al., Nat. Biotechnol. 28:157-159 (2010))
- [NPL12] Zheng et al., Clin. Pharm. & Ther. 89(2):283-290 (2011)
- [NPL13] Jefferis et al., Immunol. Lett. 82:57-65 (2002)
- [NPL14] Lazar et al., Proc. Nat. Acad. Sci. USA. 103:4005-4010 (2006)
- [NPL15] Shinkawa et al., J. Biol. Chem. 278, 3466-3473 (2003)
- [NPL16] Clynes et al., Proc. Natl. Acad. Sci. U SA 95:652-656 (1998)
- [NPL17] Clynes et al., Nat. Med. 6:443-446 (2000)
- [NPL18] Li et al., Proc. Nat. Acad. Sci. USA. 109 (27):10966-10971 (2012)
- [NPL19] Scappaticci et al., J. Natl. Cancer Inst. 99 (16):1232-1239 (2007)
- [NPL20] Desai et al., J. Immunol. 178(10):6217-6226 (2007)
- [NPL21] Holash et al., Proc. Natl. Acad. Sci., 99(17):11393-11398 (2002)
- [NPL22] Russo et al., Exp. Rev. Clin. Immunol. 10(5):593-619 (2014)
- [NPL23] Desai et al., J. Immunol. 178(10):6217-6226 (2007)

Summary

[0031] In one nonexclusive example, a non-limited objective of examples of Disclosure A is to provide molecules with improved pharmacokinetic properties over antibodies, such as ion concentration-dependent antigen binding properties that improve antibody half-life and/or antigen clearance from the plasma. In one aspect, the present disclosure refers to an antibody comprising an antigen-binding domain whose antigen-binding activity changes according to ion concentration conditions, wherein the antibody comprises a heavy chain variable region and a light chain variable region and the antibody comprises amino acid substitutions at: (a) position 16, 43, and 105 in a FR of the heavy chain variable region; (b) position 64 in a CDR of the heavy chain variable region; (c) position 18, 45, 79, and 107 in a FR of the light chain variable region; and (d) position 24 in a CDR of the light chain variable region, according to Kabat numbering, and wherein the amino acid substitutions are selected from the group consisting of: (i) the substitution of an amino acid with a negatively charged side chain with Gln (Q), (ii) the substitution of an amino acid with a negatively charged side chain with Lys (K) or Arg (R), and (iii) the substitution of an uncharged amino acid residue with Lys (K) or Arg (R).

[0032] In one nonexclusive example, a non-limited objective of examples of Disclosure B is to provide, safe and more favorable Fc region variants that have increased half-life and decreased binding to pre-existing anti-drug antibodies (ADAs).

[0033] In one nonexclusive example, a non-limited objective of examples of Disclosure C is to provide anti-IL-8 antibodies that have pH-dependent binding affinity towards IL-8. An additional example relates to anti-IL-8 antibodies that have an effect of rapidly eliminating IL-8 compared to a reference antibody when administered to an individual. In another example, Disclosure C relates to anti-IL-8 antibodies that can

stably maintain their IL-8-neutralizing activity when administered to an individual. In some examples, the anti-IL-8 antibodies display reduced immunogenicity. In additional examples, Disclosure C relates to a method of producing and using the above-mentioned anti-IL-8 antibodies. Another alternative non-limited objective of Disclosure C is, to provide novel anti-IL-8 antibodies that can be included in a pharmaceutical composition.

[0034] In one nonexclusive example, within the scope of Disclosure A as provided herein, the inventors have surprisingly discovered that the ability of an ion concentration-dependent antibody (which is an antibody comprising an ion concentration-dependent antigen-binding domain ("an antigen-binding domain whose antigen-binding activity changes according to ion concentration conditions")) to eliminate antigen from plasma can be accelerated by modifying at least one of the amino acid residues exposed on the surface of the antibody to increase its isoelectric point (pI). In another nonexclusive example, the inventors discovered that an ion concentration-dependent antibody with increased pI can further increase the extracellular matrix-binding of the antibody. Thus, without being confined to a particular theory, the inventors have discovered that antigen elimination from plasma can be increased, by increasing the binding of the antibody towards extracellular matrix.

[0035] In one nonexclusive example, within the scope of Disclosure B as provided herein, the inventors conducted dedicated research on safe and more favorable Fc region variants that do not show binding to anti-drug antibodies (pre-existing ADA) and that can further improve plasma retention. As a result, the inventors have surprisingly discovered that Fc region variants comprising a substitution of position 434 amino acid according to EU numbering with Ala (A) and two specific residue mutations (represented by Q438R/S440E according to EU numbering) as a combination of amino acid residue mutations, are preferred for maintaining significant reduction in the binding to rheumatoid factor, along with achieving a plasma retention of an antibody.

[0036] In one nonexclusive example, within the scope of Disclosure C as provided herein, the inventors generated a number of pH-dependent anti-IL-8 antibodies (anti-IL-8 antibodies that bind to IL-8 in a pH-dependent manner). From the results of various validations, the inventors identified pH-dependent anti-IL-8 antibodies that have an effect of rapidly eliminating IL-8 compared to a reference antibody when administered to an individual. In some examples the Disclosure C relates to pH-dependent anti-IL-8 antibodies that can stably maintain their IL-8-neutralizing activity. In additional nonlimiting examples, the pH-dependent anti-IL-8 antibodies have reduced immunogenicity and excellent expression levels.

[0037] Further, within the scope of Disclosure C, the inventors successfully obtained anti-IL-8 antibodies that comprise an Fc region whose FcRn-binding affinity at acidic pH is

increased relative to the FcRn-binding affinity of a native Fc region. In an alternative example, the inventors successfully obtained anti-IL-8 antibodies that comprise an Fc region whose binding affinity towards pre-existing ADA is reduced relative to the binding affinity of a native Fc region for the pre-existing ADA. In an alternative example, the inventors successfully obtained anti-IL-8 antibodies comprising an Fc region that whose plasma half-life is increased relative to the plasma half-life of a native Fc region. In an alternative example, the inventors successfully obtained pH-dependent anti-IL-8 antibodies that comprise an Fc region whose binding affinity towards effector receptors is reduced relative to the binding affinity of a naturally occurring Fc region for the effector receptors. In a different example, the inventors identified nucleic acids encoding the above-mentioned anti-IL-8 antibodies. In another example, the inventors also obtained hosts comprising the above-mentioned nucleic acids. In another example, the inventors developed a method for producing the above-mentioned anti-IL-8 antibodies, which comprises culturing the above-mentioned host. In another example, the inventors developed a method for facilitating the elimination of IL-8 from an individual relative to a reference antibody, which comprises administering the above-mentioned anti-IL-8 antibodies to the individual.

[0038] In one example, Disclosure A, relates without limitation to,

- [1] an antibody comprising an antigen-binding domain whose antigen-binding activity changes according to ion concentration conditions, wherein its isoelectric point (pI) is increased by the modification of at least one amino acid residue that may be exposed on the surface of the antibody;
- [2] the antibody of [1], wherein the antigen is a soluble antigen;
- [3] the antibody of [1] or [2], wherein the antigen-binding domain is a domain whose antigen-binding activity under a high ion concentration condition is higher than that under a low ion concentration condition;
- [4] the antibody of any one of [1] to [3], wherein the ion concentration is a hydrogen ion concentration (pH) or a calcium ion concentration;
- [5] the antibody of [4], wherein the ratio of its K_D in an acidic pH range to that in a neutral pH range, K_D (acidic pH range) / K_D (neutral pH range), for the antigen, is 2 or higher;
- [6] the antibody of any one of [1] to [5], wherein in the antigen-binding domain, at least one amino acid residue is substituted with histidine, or at least one histidine is inserted;
- [7] the antibody of any one of [1] to [6], which can promote elimination of the antigen from plasma as compared to an antibody before the modification;
- [8] the antibody of any one of [1] to [7], wherein its extracellular matrix-binding activity is enhanced as compared to an antibody before the modification;

[9] the antibody of any one of [1] to [8], wherein the amino acid residue modification is amino acid residue substitution;

[10] the antibody of any one of [1] to [9], wherein the amino acid residue modification is selected from the group consisting of:

- (a) substitution of a negatively charged amino acid residue with an uncharged amino acid residue;
- (b) substitution of a negatively charged amino acid residue with a positively charged amino acid residue; and
- (c) substitution of an uncharged amino acid residue with a positively charged amino acid residue;

[11] the antibody of any one of [1] to [10], wherein the antibody comprises a variable region and/or a constant region, and the amino acid residue modification is amino acid residue modification in the variable region and/or the constant region;

[12] the antibody of [11], wherein the variable region comprises complementarity-determining region(s) (CDR(s)) and/or framework region(s) (FR(s));

[13] the antibody of [12], wherein the variable region comprises a heavy chain variable region and/or a light chain variable region, and at least one amino acid residue is modified in a position in a CDR or a FR selected from the group consisting of:

- (a) position 1, 3, 5, 8, 10, 12, 13, 15, 16, 18, 19, 23, 25, 26, 39, 41, 42, 43, 44, 46, 68, 71, 72, 73, 75, 76, 77, 81, 82, 82a, 82b, 83, 84, 85, 86, 105, 108, 110, and 112 in a FR of the heavy chain variable region;
- (b) position 31, 61, 62, 63, 64, 65, and 97 in a CDR of the heavy chain variable region;
- (c) position 1, 3, 7, 8, 9, 11, 12, 16, 17, 18, 20, 22, 37, 38, 39, 41, 42, 43, 45, 46, 49, 57, 60, 63, 65, 66, 68, 69, 70, 74, 76, 77, 79, 80, 81, 85, 100, 103, 105, 106, 107, and 108 in a FR of the light chain variable region; and
- (d) position 24, 25, 26, 27, 52, 53, 54, 55, and 56 in a CDR of the light chain variable region, according to Kabat numbering;

[14] the antibody of [13], wherein at least one amino acid residue is modified in a position in a CDR or a FR selected from the group consisting of:

- (a) position 8, 10, 12, 13, 15, 16, 18, 23, 39, 41, 43, 44, 77, 82, 82a, 82b, 83, 84, 85, and 105 in a FR of the heavy chain variable region;
 - (b) position 31, 61, 62, 63, 64, 65, and 97 in a CDR of the heavy chain variable region;
 - (c) position 16, 18, 37, 41, 42, 45, 65, 69, 74, 76, 77, 79, and 107 in a FR of the light chain variable region; and
 - (d) position 24, 25, 26, 27, 52, 53, 54, 55, and 56 in a CDR of the light chain variable region;
- [15] the antibody of any one of [11] to [14], wherein at least one amino acid residue is modified in a position in the constant region selected from the group consisting of

- position 196, 253, 254, 256, 258, 278, 280, 281, 282, 285, 286, 307, 309, 311, 315, 327, 330, 342, 343, 345, 356, 358, 359, 361, 362, 373, 382, 384, 385, 386, 387, 389, 399, 400, 401, 402, 413, 415, 418, 419, 421, 424, 430, 433, 434, and 443, according to EU numbering;
- [16] the antibody of [15], wherein at least one amino acid residue is modified in a position in the constant region selected from the group consisting of position 254, 258, 281, 282, 285, 309, 311, 315, 327, 330, 342, 343, 345, 356, 358, 359, 361, 362, 384, 385, 386, 387, 389, 399, 400, 401, 402, 413, 418, 419, 421, 433, 434, and 443;
- [17] the antibody of [16], wherein at least one amino acid residue is modified in a position in the constant region selected from the group consisting of position 282, 309, 311, 315, 342, 343, 384, 399, 401, 402, and 413, according to EU numbering;
- [18] the antibody of any one of [1] to [17], wherein the constant region has Fc gamma receptor (FcγR)-binding activity, and wherein the FcγR-binding activity under a neutral pH condition is enhanced as compared to that of a reference antibody comprising a constant region of a native IgG;
- [19] the antibody of [18], wherein the FcγR is FcγRIIb;
- [20] the antibody of any one of [1] to [17], wherein the constant region has binding activity towards one or more activating FcγR selected from the group consisting of FcγRIa, FcγRIb, FcγRIc, FcγRIIIa, FcγRIIIb and FcγRIIa, and towards FcγRIIb, and the FcγRIIb-binding activity is maintained or enhanced and the binding activity to the activating FcγRs is decreased, as compared to those of a reference antibody which differs only in that its constant region is that of a native IgG;
- [21] the antibody of any one of [1] to [20], wherein the constant region has FcRn-binding activity, and wherein the FcRn-binding activity under a neutral pH condition (e.g., pH 7.4) is enhanced as compared to that of a reference antibody which differs only in that its constant region is that of a native IgG;
- [22] the antibody of any one of [1] to [21], which is a multispecific antibody that binds to at least two antigens;
- [23] the antibody of any one of [1] to [22], wherein the antibody is an IgG antibody;
- [24] a pharmaceutical composition comprising the antibody of any one of [1] to [23];
- [25] the pharmaceutical composition of [24], which is for promoting the elimination of an antigen from plasma;
- [26] the pharmaceutical composition of [24] or [25], which is for enhancing the antibody binding to an extracellular matrix;
- [27] a nucleic acid encoding the antibody of any one of [1] to [23];
- [28] a vector comprising the nucleic acid of [27];
- [29] a host cell comprising the vector of [28];
- [30] a method for producing an antibody comprising an antigen-binding domain whose

antigen-binding activity changes according to ion concentration conditions, wherein the method comprises culturing the host cell of [29] and collecting the antibody from the cell culture;

[30A] a method for producing an antibody comprising an antigen-binding domain whose antigen-binding activity changes according to ion concentration conditions, wherein the method comprises modifying at least one amino acid residue that may be exposed on the surface of the antibody so as to increase the isoelectric point (pI);

[30B] the method of [30A], wherein at least one amino acid residue is modified

(I) in a position in a CDR or FR selected from the group consisting of: (a) position 1, 3, 5, 8, 10, 12, 13, 15, 16, 18, 19, 23, 25, 26, 39, 41, 42, 43, 44, 46, 68, 71, 72, 73, 75, 76, 77, 81, 82, 82a, 82b, 83, 84, 85, 86, 105, 108, 110, and 112 in a FR of the heavy chain variable region; (b) position 31, 61, 62, 63, 64, 65, and 97 in a CDR of the heavy chain variable region; (c) position 1, 3, 7, 8, 9, 11, 12, 16, 17, 18, 20, 22, 37, 38, 39, 41, 42, 43, 45, 46, 49, 57, 60, 63, 65, 66, 68, 69, 70, 74, 76, 77, 79, 80, 81, 85, 100, 103, 105, 106, 107, and 108 in a FR of the light chain variable region; and (d) position 24, 25, 26, 27, 52, 53, 54, 55, and 56 in a CDR of the light chain variable region, according to Kabat numbering; or

(II) in a position in a constant region selected from the group consisting of position 196, 253, 254, 256, 258, 278, 280, 281, 282, 285, 286, 307, 309, 311, 315, 327, 330, 342, 343, 345, 356, 358, 359, 361, 362, 373, 382, 384, 385, 386, 387, 389, 399, 400, 401, 402, 413, 415, 418, 419, 421, 424, 430, 433, 434, and 443, according to EU numbering;

[31] the method of [30A] or [30B], wherein the amino acid residue modification comprises a modification selected from the group consisting of:

(a) substitution of a negatively charged amino acid residue with an uncharged amino acid residue;

(b) substitution of a negatively charged amino acid residue with a positively charged amino acid residue;

(c) substitution of an uncharged amino acid residue with a positively charged amino acid residue; and

(d) substitution or insertion with histidine in a CDR or FR.

[32] the method of any one of [30], or [30A] to [30C] which further optionally comprises any one or more of:

(a) selecting an antibody which can promote elimination of an antigen from plasma;

(b) selecting an antibody with enhanced binding activity to an extracellular matrix;

(c) selecting an antibody with enhanced FcγR-binding activity under a neutral pH condition (e.g., pH 7.4);

(d) selecting an antibody with enhanced FcγRIIb-binding activity under a neutral pH

condition (e.g., pH 7.4);

(e) selecting an antibody with maintained or enhanced FcγRIIb-binding activity and decreased binding activity to one or more activating FcγR, preferably selected from the group consisting of FcγRIa, FcγRIb, FcγRIc, FcγRIIIa, FcγRIIIb and FcγRIIa;

(f) selecting an antibody with enhanced FcRn-binding activity under a neutral pH condition (e.g., pH 7.4);

(g) selecting an antibody with an increased isoelectric point (pI);

(h) confirming the isoelectric point (pI) of the collected antibody, and then selecting an antibody with an increased isoelectric point (pI); and

(i) selecting an antibody whose antigen-binding activity is changed or increased according to ion concentration conditions;

as compared to a reference antibody;

[0039] In an alternative example, Disclosure A relates without limitation to:

[A1] an antibody having a constant region, wherein at least one amino acid residue selected from the group of modification sites identical to the modification sites in the group defined in [15] or [16] is modified in the constant region;

[A2] the antibody of [A1], which further has a heavy-chain variable region and/or a light-chain variable region, wherein the variable region has CDR(s) and/or FR(s), and wherein at least one amino acid residue selected from the group of modification sites identical to the modification sites in the group defined in [13] or [14] is modified in a CDR and/or a FR;

[A3] an antibody having a constant region, wherein at least one amino acid residue selected from the group of modification sites identical to the modification sites in the group defined in [15] or [16] is modified in the constant region so as to increase its pI;

[A4] the antibody of [A3], which further has a heavy-chain variable region and/or a light-chain variable region, wherein the variable region has CDR(s) and/or FR(s), and wherein at least one amino acid residue selected from the group of modification sites identical to the modification sites in the group defined in [13] or [14] is modified in a CDR and/or a FR;

[A5] an antibody comprising an antigen-binding domain whose antigen-binding activity changes according to ion concentration conditions, wherein the antibody has a constant region, and wherein at least one amino acid residue selected from the group of modification sites identical to the modification sites in the group defined in [15] or [16] is modified in the constant region;

[A6] the antibody of [A5], which further has a heavy-chain variable region and/or a light-chain variable region, wherein the variable region has CDR(s) and/or FR(s), and wherein at least one amino acid residue selected from the group of modification sites identical to the modification sites in the group defined in [13] or [14] is modified in a

CDR and/or a FR;

[A7] use of the antibody of any one of [1] to [23] and [A1] to [A6] in the manufacture of a medicament for promoting antigen elimination from plasma;

[A8] use of the antibody of any one of [1] to [23] and [A1] to [A6] in the manufacture of a medicament for increasing extracellular matrix binding;

[A9] use of the antibody of any one of [1] to [23] and [A1] to [A6] for eliminating an antigen from plasma; and

[A10] use of the antibody of any one of [1] to [23] and [A1] to [A6] for increasing extracellular matrix binding.

[A11] an antibody obtained by the method of any one of [30], [30A], [30B], [31], [32].

[0040] According to various examples, Disclosure A encompasses combinations of one or multiple elements described in any of [1] to [30], [30A], [30B], [31], [32] and [A1] to [A11] mentioned above, in part or as a whole, as long as such a combination is not technically inconsistent with the common technical knowledge in the art. For example, in some examples, Disclosure A encompasses a method for producing a modified antibody comprising an antigen-binding domain which promotes elimination of an antigen from plasma as compared to that before the antibody modification, wherein the method comprises:

(a) modifying at least one amino acid residue that may be exposed on the surface of an antibody, which is:

(I) in a position in a CDR or FR selected from the group consisting of: (a) position 1, 3, 5, 8, 10, 12, 13, 15, 16, 18, 19, 23, 25, 26, 39, 41, 42, 43, 44, 46, 68, 71, 72, 73, 75, 76, 77, 81, 82, 82a, 82b, 83, 84, 85, 86, 105, 108, 110, and 112 in a FR of the heavy chain variable region;

(b) position 31, 61, 62, 63, 64, 65, and 97 in a CDR of the heavy chain variable region; (c) position 1, 3, 7, 8, 9, 11, 12, 16, 17, 18, 20, 22, 37, 38, 39, 41, 42, 43, 45, 46, 49, 57, 60, 63, 65, 66, 68, 69, 70, 74, 76, 77, 79, 80, 81, 85, 100, 103, 105, 106, 107, and 108 in a FR of the light chain variable region; and (d) position 24, 25, 26, 27, 52, 53, 54, 55, and 56 in a CDR of the light chain variable region, according to Kabat numbering; or

(II) in a position in a constant region selected from the group consisting of position 196, 253, 254, 256, 258, 278, 280, 281, 282, 285, 286, 307, 309, 311, 315, 327, 330, 342, 343, 345, 356, 358, 359, 361, 362, 373, 382, 384, 385, 386, 387, 389, 399, 400, 401, 402, 413, 415, 418, 419, 421, 424, 430, 433, 434, and 443, according to EU numbering;

(b) modifying the antigen-binding domain in a way such that the resulting antigen-binding activity changes according to ion concentration conditions, wherein said (a) and (b) can be carried out simultaneously or sequentially;

(c) culturing a host cell to express the nucleic acid encoding the modified antibody;

and

(d) collecting the modified antibody from the host cell culture.

In further examples, the method optionally further comprises any one or more of:

(e) selecting an antibody which can promote elimination of an antigen from plasma;

(f) selecting an antibody with enhanced binding activity to an extracellular matrix;

(g) selecting an antibody with enhanced FcγR-binding activity under a neutral pH condition (e.g., pH 7.4);

(h) selecting an antibody with enhanced FcγRIIb-binding activity under a neutral pH condition (e.g., pH 7.4);

(i) selecting an antibody with maintained or enhanced FcγRIIb-binding activity and decreased binding activity to one or more activating FcγR, preferably selected from the group consisting of FcγRIa, FcγRIb, FcγRIc, FcγRIIIa, FcγRIIIb and FcγRIIa;

(j) selecting an antibody with enhanced FcRn-binding activity under a neutral pH condition (e.g., pH 7.4);

(k) selecting an antibody with an increased isoelectric point (pI);

(l) confirming the isoelectric point (pI) of the collected antibody, and then selecting an antibody with an increased isoelectric point (pI); and

(m) selecting an antibody whose antigen-binding activity is changed or increased according to ion concentration conditions;

as compared to the antibody before the modification.

[0040a] In another aspect, the present disclosure refers to a pharmaceutical composition comprising the antibody as described herein. In yet another aspect, the present disclosure refers to a nucleic acid encoding the antibody as described herein. In a still further aspect, the present disclosure refers to a vector comprising the nucleic acid as described herein. In another aspect, the present disclosure refers to a host cell comprising the vector as described herein. In yet a further aspect, the present disclosure refers to a method for producing an antibody comprising an antigen-binding domain whose antigen-binding activity changes according to ion concentration conditions, wherein the method comprises culturing the host cell as described herein and collecting the antibody from the cell culture.

[0041] Another example of Disclosure A relates to, for example, without limitation:

[D1] a method for producing a modified antibody, whose half-life in plasma is prolonged or reduced, as compared to that before the modification of the antibody, wherein the method comprises:

(a) modifying a nucleic acid encoding the antibody before the modification to change the charge of at least one amino acid residue at a position selected from the group consisting of position 196, 253, 254, 256, 258, 278, 280, 281, 282, 285, 286, 307, 309, 311, 315, 327, 330, 342, 343, 345, 356, 358, 359, 361, 362, 373, 382, 384, 385, 386, 387, 389, 399, 400, 401, 402, 413, 415, 418, 419, 421, 424, 430, 433, 434, and 443, according to EU numbering;

(b) culturing a host cell to express the nucleic acid; and

(c) collecting the antibody from the host cell culture; or

[D2] a method for prolonging or reducing the half-life of an antibody in plasma wherein the method comprises modifying at least one amino acid residue at a position selected from the group consisting of position 196, 253, 254, 256, 258, 278, 280, 281, 282, 285, 286, 307, 309, 311, 315, 327, 330, 342, 343, 345, 356, 358, 359, 361, 362, 373, 382, 384, 385, 386, 387, 389, 399, 400, 401, 402, 413, 415, 418, 419, 421, 424, 430, 433, 434, and 443, according to EU numbering.

[0042] In one example, Disclosure B relates to, for example, without limitation:

[33] an Fc region variant comprising an FcRn-binding domain, wherein the FcRn-binding domain comprises Ala at position 434; Glu, Arg, Ser, or Lys at position 438; and Glu, Asp, or Gln at position 440, according to EU numbering;

[34] the Fc region variant of [33], wherein the FcRn-binding domain comprises Ala at position 434; Arg or Lys at position 438; and Glu or Asp at position 440, according to EU numbering;

[35] the Fc region variant of [33] or [34], wherein the FcRn-binding domain further comprises Ile or Leu at position 428; and/or Ile, Leu, Val, Thr, or Phe at position 436, according to EU numbering;

[36] the Fc region variant of [35], wherein the FcRn-binding domain comprises Leu at position 428; and/or Val or Thr at position 436, according to EU numbering;

[37] the Fc region variant of any one of [33] to [36], wherein the FcRn-binding domain comprises a combination of amino acid substitutions selected from the group consisting of:

N434A/Q438R/S440E; N434A/Q438R/S440D; N434A/Q438K/S440E;
 N434A/Q438K/S440D; N434A/Y436T/Q438R/S440E; N434A/Y436T/Q438R/S440D;
 N434A/Y436T/Q438K/S440E; N434A/Y436T/Q438K/S440D; N434A/Y436V/Q438R/
 S440E; N434A/Y436V/Q438R/S440D; N434A/Y436V/Q438K/S440E;
 N434A/Y436V/Q438K/S440D; N434A/R435H/F436T/Q438R/S440E;
 N434A/R435H/F436T/Q438R/S440D; N434A/R435H/F436T/Q438K/S440E;
 N434A/R435H/F436T/Q438K/S440D; N434A/R435H/F436V/Q438R/S440E;
 N434A/R435H/F436V/Q438R/S440D; N434A/R435H/F436V/Q438K/S440E;
 N434A/R435H/F436V/Q438K/S440D; M428L/N434A/Q438R/S440E;
 M428L/N434A/Q438R/S440D; M428L/N434A/Q438K/S440E;
 M428L/N434A/Q438K/S440D; M428L/N434A/Y436T/Q438R/S440E;
 M428L/N434A/Y436T/Q438R/S440D; M428L/N434A/Y436T/Q438K/S440E;
 M428L/N434A/Y436T/Q438K/S440D; M428L/N434A/Y436V/Q438R/S440E;
 M428L/N434A/Y436V/Q438R/S440D; M428L/N434A/Y436V/Q438K/S440E;
 M428L/N434A/Y436V/Q438K/S440D;

L235R/G236R/S239K/M428L/N434A/Y436T/Q438R/S440E; and

L235R/G236R/A327G/A330S/P331S/M428L/N434A/Y436T/Q438R/S440E, according to EU numbering;

[38] the Fc region variant of [37], wherein the FcRn-binding domain comprises a combination of amino acid substitutions selected from the group consisting of:

N434A/Q438R/S440E; N434A/Y436T/Q438R/S440E; N434A/Y436V/Q438R/S440E;
 M428L/N434A/Q438R/S440E; M428L/N434A/Y436T/Q438R/S440E;
 M428L/N434A/Y436V/Q438R/S440E;

L235R/G236R/S239K/M428L/N434A/Y436T/Q438R/S440E; and

L235R/G236R/A327G/A330S/P331S/M428L/N434A/Y436T/Q438R/S440E, according to EU numbering;

[39] the Fc region variant of any one of [33] to [38], wherein its FcRn-binding activity under an acidic pH condition (e.g., pH 5.8) is enhanced as compared to that of an Fc region of a native IgG;

[40] the Fc region variant of any one of [33] to [39], wherein its binding activity to an anti-drug antibody (ADA) is not significantly enhanced under a neutral pH condition as compared to that of an Fc region of a native IgG;

[41] the Fc region variant of [40], wherein the anti-drug antibody (ADA) is a rheumatoid factor (RF);

[42] the Fc region variant of any one of [33] to [41], wherein its plasma clearance (CL) is decreased, plasma retention time is increased, or plasma half-life ($t_{1/2}$) is increased, as compared to that of an Fc region of a native IgG;

[43] the Fc region variant of any one of [33] to [42], wherein its plasma retention is increased as compared to a reference Fc region variant comprising a combination of amino acid substitutions N434Y/Y436V/Q438R/S440E, according to EU numbering;

[44] an antibody comprising the Fc region variant of any one of [33] to [43];

[45] the antibody of [44], wherein the antibody is an IgG antibody;

[46] a pharmaceutical composition comprising the antibody of [44] or [45];

[47] the pharmaceutical composition of [46], which is for increasing retention of the antibody in plasma;

[48] a nucleic acid encoding the Fc region variant of any one of [33] to [43] or the antibody of [44] or [45];

[49] a vector comprising the nucleic acid of [48];

[50] a host cell comprising the vector of [49];

[51] a method for producing an Fc region variant comprising an FcRn-binding domain or an antibody comprising the variant, which comprises culturing the host cell of [50], and then collecting the Fc region variant or the antibody comprising the variant from the cell culture;

[52] the method of [51], which further optionally comprises any one or more steps selected from the group consisting of:

(a) selecting an Fc region variant with enhanced FcRn-binding activity under an acidic pH condition as compared to that of an Fc region of a native IgG;

(b) selecting an Fc region variant whose binding activity to an anti-drug antibody (ADA) is not significantly enhanced under a neutral pH condition as compared to that of an Fc region of a native IgG;

(c) selecting an Fc region variant with increased plasma retention as compared to that

of an Fc region of a native IgG; and

(d) selecting an antibody comprising an Fc region variant that can promote elimination of an antigen from plasma as compared to a reference antibody comprising an Fc region of a native IgG; and

[53] a method for producing an Fc region variant comprising an FcRn-binding domain or an antibody comprising the variant, wherein the method comprises substituting amino acids in a way such that the resulting Fc region variant or the antibody comprising the variant comprises Ala at position 434; Glu, Arg, Ser, or Lys at position 438; and Glu, Asp, or Gln at position 440, according to EU numbering.

[0043] In one example, Disclosure B relates to, for example, without limitation:

[B1] use of the Fc region variant of any one of [33] to [43] or the antibody of [44] or [45] in the manufacture of a medicament for increasing retention in plasma;

[B2] use of the Fc region variant of any one of [33] to [43] or the antibody of [44] or [45] in the manufacture of a medicament for not significantly increasing the binding activity for an anti-drug antibody (ADA) under a neutral pH condition compared to the Fc region of a native IgG;

[B3] use of the Fc region variant of any one of [33] to [43] or the antibody of [44] or [45] for increasing retention in plasma;

[B4] use of the Fc region variant of any one of [33] to [43] or the antibody of [44] or [45] for not significantly increasing the binding activity for an anti-drug antibody (ADA) under a neutral pH condition compared to the Fc region of a native IgG; and

[B5] an Fc region variant or an antibody comprising the variant, which is obtained by the method of any one of [51], [52], and [53].

[0044] According to various examples, Disclosure B encompasses combinations of one or multiple elements described in any of [33] to [53] and [B1] to [B5] mentioned above, in part or as a whole, as long as such a combination is not technically inconsistent with the common technical knowledge in the art. For example, in some examples, Disclosure B encompasses an Fc region variant comprising an FcRn-binding domain, wherein the FcRn-binding domain can comprise:

(a) Ala at position 434; Glu, Arg, Ser, or Lys at position 438; and Glu, Asp, or Gln at position 440, according to EU numbering;

(b) Ala at position 434; Arg or Lys at position 438; and Glu or Asp at position 440, according to EU numbering;

(c) Ile or Leu at position 428; Ala at position 434; Ile, Leu, Val, Thr, or Phe at position 436; Glu, Arg, Ser, or Lys at position 438; and Glu, Asp, or Gln at position 440, according to EU numbering;

(d) Ile or Leu at position 428; Ala at position 434; Ile, Leu, Val, Thr, or Phe at position 436; Arg or Lys at position 438; and Glu or Asp at position 440, according to

EU numbering;

(e) Leu at position 428; Ala at position 434; Val or Thr at position 436; Glu, Arg, Ser, or Lys at position 438; and Glu, Asp, or Gln at position 440, according to EU numbering; or

(f) Leu at position 428; Ala at position 434; Val or Thr at position 436; Arg or Lys at position 438; and Glu or Asp at position 440, according to EU numbering.

[0045]

In one example, Disclosure C relates to, for example, without limitation:

[54] an isolated anti-IL-8 antibody that binds to human IL-8, which comprises at least one amino acid substitution(s) in at least one of (a) to (f) below, and binds to IL-8 in a pH-dependent manner:

(a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:67;

(b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:68;

(c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:69;

(d) HVR-L1 comprising the amino acid sequence of SEQ ID NO:70;

(e) HVR-L2 comprising the amino acid sequence of SEQ ID NO:71; and

(f) HVR-L3 comprising the amino acid sequence of SEQ ID NO:72;

[55] the anti-IL-8 antibody of [54], which comprises an amino acid substitutions of tyrosine at position 9 of the amino acid sequence of SEQ ID NO:68, arginine at position 11 of the amino acid sequence of SEQ ID NO:68, and tyrosine at position 3 of the amino acid sequence of SEQ ID NO:69;

[56] the anti-IL-8 antibody of [54] or [55], which further comprises an amino acid substitutions of alanine at position 6 of the amino acid sequence of SEQ ID NO:68 and glycine at position 8 of the amino acid sequence of SEQ ID NO:68;

[57] the anti-IL-8 antibody of any one of [54] to [56], which comprises an amino acid substitutions of asparagine at position 1 of the amino acid sequence of SEQ ID NO:71, leucine at position 5 of the amino acid sequence of SEQ ID NO:71, and glutamine at position 1 of the amino acid sequence of SEQ ID NO:72;

[58] the anti-IL-8 antibody of any one of [54] to [57], which comprises (a) HVR-H1 comprising the amino acid sequence of SEQ ID NO:67, (b) HVR-H2 comprising the amino acid sequence of SEQ ID NO:73, and (c) HVR-H3 comprising the amino acid sequence of SEQ ID NO:74;

[59] the anti-IL-8 antibody of any one of [54] to [58], which comprises (a) HVR-L1 comprising the amino acid sequence of SEQ ID NO:70, (b) HVR-L2 comprising the amino acid sequence of SEQ ID NO:75, and (c) HVR-L3 comprising the amino acid sequence of SEQ ID NO:76;

[60] the anti-IL-8 antibody of any one of [54] to [59], which comprises the heavy chain variable region of SEQ ID NO:78 and the light chain variable region of SEQ ID NO:79;

[61] the anti-IL-8 antibody of any one of [54] to [60], which comprises an Fc region having at least one property selected from the properties of (a) to (f) below:

- (a) increased binding affinity for FcRn of the Fc region relative to the binding affinity for FcRn of a native Fc region at acidic pH;
- (b) reduced binding affinity of the Fc region for pre-existing ADA relative to the binding affinity of a native Fc region for the pre-existing ADA;
- (c) increased plasma half-life of the Fc region relative to the plasma half-life of a native Fc region;
- (d) reduced plasma clearance of the Fc region relative to the plasma clearance of a native Fc region; and
- (e) reduced binding affinity of the Fc region for an effector receptor relative to the binding affinity of a native Fc region for the effector receptor; and
- (f) increased binding to extracellular matrix.

[62] the anti-IL-8 antibody of [61], wherein the Fc region comprises amino acid substitution(s) at one or more positions selected from the group consisting of position 235, 236, 239, 327, 330, 331, 428, 434, 436, 438 and 440, according to EU numbering;

[63] the anti-IL-8 antibody of [62], which comprises an Fc region comprising one or more amino acid substitutions selected from the group consisting of L235R, G236R, S239K, A327G, A330S, P331S, M428L, N434A, Y436T, Q438R and S440E;

[64] the anti-IL-8 antibody of [63], wherein the Fc region comprises the amino acid substitutions of L235R, G236R, S239K, M428L, N434A, Y436T, Q438R and S440E;

[65] the anti-IL-8 antibody of [63], wherein the Fc region comprises the amino acid substitution of L235R, G236R, A327G, A330S, P331S, M428L, N434A, Y436T, Q438R and S440E;

[66] an anti-IL-8 antibody that comprises a heavy chain comprising the amino acid sequence of SEQ ID NO:81 and a light chain comprising the amino acid sequence of SEQ ID NO:82;

[67] an anti-IL-8 antibody that comprises a heavy chain comprising the amino acid sequence of SEQ ID NO:80 and a light chain comprising the amino acid sequence of SEQ ID NO:82;

[68] an isolated nucleic acid encoding the anti-IL-8 antibody of any one of [54] to [67];

[69] a vector comprising the nucleic acid of [68];

[70] a host cell comprising the vector of [69];

[71] a method for producing an anti-IL-8 antibody, which comprises culturing the host of [70];

[72] the method for producing an anti-IL-8 antibody of [71], which comprises isolating the antibody from a culture supernatant;

[73] a pharmaceutical composition comprising the anti-IL-8 antibody of any one of

- [54] to [67], and a pharmaceutically acceptable carrier;
- [74] the anti-IL-8 antibody of any one of [54] to [67] for use in a pharmaceutical composition;
- [75] the anti-IL-8 antibody of any one of [54] to [67] for use in the treatment of a disorder with the presence of excess IL-8;
- [76] use of the anti-IL-8 antibody of any one of [54] to [67] in the manufacture of a pharmaceutical composition for a disorder with the presence of excess IL-8;
- [77] a method for treating a patient that has a disorder with the presence of excess IL-8, which comprises administering the anti-IL-8 antibody of any one of [54] to [67] to the individual;
- [78] a method for promoting elimination of IL-8 from an individual, which comprises administering the anti-IL-8 antibody of any one of [54] to [67] to the individual;
- [79] a pharmaceutical composition comprising the anti-IL-8 antibody of any one of [54] to [67], wherein the antibody binds to IL-8 and binds to extracellular matrix; and
- [80] a method for producing an anti-IL-8 antibody comprising a variable region with a pH-dependent IL-8-binding activity, wherein the method comprises:
- (a) evaluating binding of an anti-IL-8 antibody with extracellular matrix,
 - (b) selecting an anti-IL-8 antibody with strong binding to the extracellular matrix,
 - (c) culturing a host that comprises a vector comprising a nucleic acid encoding the antibody, and
 - (d) isolating the antibody from the culture solution.
- [0046] In an alternative example, Disclosure C relates to:
- [C1] use of the anti-IL-8 antibody of any one of [54] to [67] in the manufacture of a pharmaceutical composition for suppressing accumulation of IL-8 which has a biological activity;
- [C2] use of the anti-IL-8 antibody of any one of [54] to [67] for suppressing accumulation of IL-8 which has a biological activity;
- [C3] use of the anti-IL-8 antibody of any one of [54] to [67] in the manufacture of a pharmaceutical composition for inhibiting angiogenesis;
- [C4] use of the anti-IL-8 antibody of any one of [54] to [67] for inhibiting angiogenesis;
- [C5] use of the anti-IL-8 antibody of any one of [54] to [67] in the manufacture of a pharmaceutical composition for inhibiting facilitation of neutrophil migration;
- [C6] use of the anti-IL-8 antibody of any one of [54] to [67] for inhibiting facilitation of neutrophil migration;
- [C7] the anti-IL-8 antibody of any one of [54] to [67] for use in suppressing accumulation of IL-8 which has a biological activity;
- [C8] a method for suppressing accumulation of IL-8 which has a biological activity,

wherein the method comprises administering the anti-IL-8 antibody of any one of [54] to [67] to an individual;

[C9] a pharmaceutical composition for suppressing accumulation of IL-8 which has a biological activity, comprising the anti-IL-8 antibody of any one of [54] to [67];

[C10] the anti-IL-8 antibody of any one of [54] to [67] for use in inhibiting angiogenesis;

[C11] a method for inhibiting angiogenesis in an individual, wherein the method comprises administering the anti-IL-8 antibody of any one of [54] to [67] to the individual;

[C12] a pharmaceutical composition for inhibiting angiogenesis, which comprises the anti-IL-8 antibody of any one of [54] to [67];

[C13] the anti-IL-8 antibody of any one of [54] to [67] for use in inhibiting facilitation of neutrophil migration;

[C14] a method for inhibiting facilitation of neutrophil migration in an individual, wherein the method comprises administering the anti-IL-8 antibody of any one of [54] to [67] to the individual;

[C15] a pharmaceutical composition for inhibiting facilitation of neutrophil migration, which comprises the anti-IL-8 antibody of any one of [54] to [67];

[C16] the anti-IL-8 antibody of any one of [54] to [67] for use in the treatment of a disorder with the presence of excess IL-8;

[C17] use of the anti-IL-8 antibody of any one of [54] to [67] in the manufacture of a pharmaceutical composition for treating a disorder with the presence of excess IL-8;

[C18] use of the anti-IL-8 antibody of any one of [54] to [67] for treating a disorder with the presence of excess IL-8;

[C19] a method for treating a disorder with the presence of excess IL-8 in an individual, wherein the method comprises administering the anti-IL-8 antibody of any one of [54] to [67] to the individual;

[C20] a pharmaceutical composition for treating a disorder with the presence of excess IL-8, which comprises the anti-IL-8 antibody of any one of [54] to [67];

[C21] the anti-IL-8 antibody of any one of [54] to [67] for use in promoting elimination of IL-8;

[C22] use of the anti-IL-8 antibody of any one of [54] to [67] in the manufacture of a pharmaceutical composition for promoting elimination of IL-8;

[C23] use of the anti-IL-8 antibody of any one of [54] to [67] for promoting elimination of IL-8;

[C24] a method for promoting elimination of IL-8 in an individual, wherein the method comprises administering the anti-IL-8 antibody of any one of [54] to [67] to the individual;
and

[C25] a pharmaceutical composition for promoting elimination of IL-8, which comprises the anti-IL-8 antibody of any one of [54] to [67].

[C26] An anti-IL-8 antibody, which comprises an Fc region comprising amino acid substitution(s) at one or more positions selected from the group consisting of position 235, 236, 239, 327, 330, 331, 428, 434, 436, 438 and 440, according to EU numbering.

[C27] The anti-IL-8 antibody of [C26], which comprises an Fc region having at least one property selected from the properties of (a) to (f) below:

- (a) increased binding affinity for FcRn of the Fc region relative to the binding affinity for FcRn of a native Fc region at acidic pH;
- (b) reduced binding affinity of the Fc region for pre-existing ADA relative to the binding affinity of a native Fc region for the pre-existing ADA;
- (c) increased plasma half-life of the Fc region relative to the plasma half-life of a native Fc region;
- (d) reduced plasma clearance of the Fc region relative to the plasma clearance of a native Fc region;
- (e) reduced binding affinity of the Fc region for an effector receptor relative to the binding affinity of a native Fc region for the effector receptor; and
- (f) increased binding to extracellular matrix.

[C28] The anti-IL-8 antibody of [C26] or [C27], which comprises an Fc region comprising one or more amino acid substitution(s) selected from the group consisting of L235R, G236R, S239K, A327G, A330S, P331S, M428L, N434A, Y436T, Q438R and S440E, according to EU numbering.

[C29] The anti-IL-8 antibody of [C28], which comprises an Fc region comprising one or more amino acid substitutions selected from the group consisting of (a) L235R, G236R, S239K, M428L, N434A, Y436T, Q438R and S440E; or (b) L235R, G236R, A327G, A330S, P331S, M428L, N434A, Y436T, Q438R and S440E, according to EU numbering.

[C30] The anti-IL-8 antibody of [C26] that comprises a heavy chain comprising the amino acid sequence of SEQ ID NO:81 and a light chain comprising the amino acid sequence of SEQ ID NO:82.

[C31] The anti-IL-8 antibody of [C26] that comprises a heavy chain comprising the amino acid sequence of SEQ ID NO:80 and a light chain comprising the amino acid sequence of SEQ ID NO:82.

[C32] An isolated nucleic acid encoding the anti-IL-8 antibody of any one of [C26] to [C31].

[C33] A vector comprising the nucleic acid of [C32].

[C34] A host cell comprising the vector of [C33].

[C35] A method for producing an anti-IL-8 antibody, which comprises culturing the

host cell of [C34].

[C36] The method for producing an anti-IL-8 antibody of any one of [C26] to [C31], which further comprises isolating the antibody from the host cell culture.

[C37] A pharmaceutical composition comprising the anti-IL-8 antibody of any one of [C26] to [C31] and a pharmaceutically acceptable carrier.

[C38] A method for treating a patient that has a disorder with the presence of excess IL-8, which comprises administering the anti-IL-8 antibody of any one of [C26] to [C31] to the individual.

[C39] A method for promoting elimination of IL-8 from an individual, which comprises administering the anti-IL-8 antibody of any one of [C26] to [C31] to the individual.

[0047] According to various examples, Disclosure C encompasses combinations of one or multiple elements described in any of [54] to [80] and [C1] to [C39] mentioned above, in part or as a whole, as long as such a combination is not technically inconsistent with the common technical knowledge in the art.

[Brief Description of Drawings]

[0048] [Fig. 1] Fig. 1 shows changes in the plasma concentration of human IL-6 receptor in human FcRn transgenic mice administered with a human IL-6 receptor-binding antibody that binds to human IL-6 receptor in a pH-dependent manner and whose constant region is that of a native IgG1 (Low_pI-IgG1), or an antibody that has increased the pI of the variable region in the antibody (High_pI-IgG1).

[0049] [Fig. 2] Fig. 2 shows changes in the plasma concentration of human IL-6 receptor in human FcRn transgenic mice administered individually with a human IL-6 receptor-binding antibody that binds to human IL-6 receptor in a pH-dependent manner and has been conferred with binding to FcRn under a neutral pH condition (Low_pI-F939), and antibodies that have increased the pI of the variable region in the antibody (Middle_pI-F939, High_pI-F939).

[0050] [Fig. 3] Fig. 3 shows changes in the plasma concentration of human IL-6 receptor in human FcRn transgenic mice administered individually with a human IL-6 receptor-binding antibody that binds to human IL-6 receptor in a pH-dependent manner and whose FcγR binding under a neutral pH condition is increased (Low_pI-F1180), and antibodies that have increased the pI of the variable region in the antibody (Middle_pI-F1180, High_pI-F1180).

[0051] [Fig. 4] Fig. 4 shows changes in the plasma concentration of human IL-6 receptor in human FcRn transgenic mice whose soluble human IL-6 receptor concentration in plasma is maintained at a steady state, which have been administered individually with a human IL-6 receptor-binding antibody that binds to human IL-6 receptor in a pH-

[CLAIMS]

[Claim 1]

An antibody comprising an antigen-binding domain whose antigen-binding activity changes according to ion concentration conditions, wherein the antibody comprises a heavy chain variable region and a light chain variable region and the antibody comprises amino acid substitutions at:

- (a) position 16, 43, and 105 in a FR of the heavy chain variable region;
 - (b) position 64 in a CDR of the heavy chain variable region;
 - (c) position 18, 45, 79, and 107 in a FR of the light chain variable region; and
 - (d) position 24 in a CDR of the light chain variable region,
- according to Kabat numbering, and wherein the amino acid substitutions are selected from the group consisting of:
- (i) the substitution of an amino acid with a negatively charged side chain with Gln (Q),
 - (ii) the substitution of an amino acid with a negatively charged side chain with Lys (K) or Arg (R), and
 - (iii) the substitution of an uncharged amino acid residue with Lys (K) or Arg (R).

[Claim 2]

The antibody of claim 1, wherein the antibody binds to a soluble antigen.

[Claim 3]

The antibody of claim 1 or 2, wherein the ion concentration is a hydrogen ion concentration (pH) or a calcium ion concentration.

[Claim 4]

The antibody of claim 43, wherein a ratio of its KD in an acidic pH range to that in a neutral pH range, $KD(\text{acidic pH range}) / KD(\text{neutral pH range})$, for the antigen, is 2 or higher.

[Claim 5]

The antibody of any one of claims 1 to 4, which can promote elimination of an antigen from plasma as compared to an antibody before the modification, or wherein an extracellular matrix-binding activity of the antibody is enhanced as compared to an antibody before the modification.

[Claim 6]

The antibody of any one of claims 1 to 5, wherein the antibody comprises a constant region.

[Claim 7]

The antibody of claim 6, wherein at least one amino acid residue is modified in a position in the constant region selected from the group consisting of

(a) position 196, 253, 254, 256, 258, 278, 280, 281, 282, 285, 286, 307, 309, 311, 315, 327, 330, 342, 343, 345, 356, 358, 359, 361, 362, 373, 382, 384, 385, 386, 387, 389, 399, 400, 401, 402, 413, 415, 418, 419, 421, 424, 430, 433, 434, and 443, according to EU numbering;

(b) position 254, 258, 281, 282, 285, 309, 311, 315, 327, 330, 342, 343, 345, 356, 358, 359, 361, 362, 384, 385, 386, 387, 389, 399, 400, 401, 402, 413, 418, 419, 421, 433, 434, and 443, according to EU numbering; or

(c) position 282, 309, 311, 315, 342, 343, 384, 399, 401, 402, and 413, according to EU numbering.

[Claim 8]

The antibody of claim 6 or 7, wherein the constant region has Fc gamma receptor (FcγR)-binding activity, and wherein the FcγR-binding activity under a neutral pH condition is enhanced as compared to that of a reference antibody comprising a constant region of a native IgG, wherein the FcγR is FcγRIIb.

[Claim 9]

The antibody of any one of claims 6 to 8, wherein the constant region has binding activity towards one or more activating FcγR selected from the group consisting of FcγRIa, FcγRIb, FcγRIc, FcγRIIIa, FcγRIIIb and FcγRIIa, and towards FcγRIIb, and the FcγRIIb-binding activity is maintained or enhanced and the binding activity to the activating FcγRs is decreased, as compared to those of a reference antibody which differs only in that its constant region is that of a native IgG.

[Claim 10]

The antibody of any one of claims 6 to 9, wherein the constant region has FcRn-binding activity, and wherein the FcRn-binding activity of the constant region under a neutral pH condition is enhanced as compared to that of a reference antibody which differs only in that its constant region is that of a native IgG.

[Claim 11]

The antibody of any one of claims 1 to 10, wherein the antibody is an IgG antibody or a multispecific antibody that binds to at least two antigens.

[Claim 12]

A pharmaceutical composition comprising the antibody of any one of claims 1 to 11.

[Claim 13]

The pharmaceutical composition of claim 12, which is for promoting elimination of an antigen from plasma or which is for enhancing binding to an extracellular matrix.

[Claim 14]

A nucleic acid encoding the antibody of any one of claims 1 to 11.

[Claim 15]

A vector comprising the nucleic acid of claim 14.

[Claim 16]

A host cell comprising the vector of claim 15.

[Claim 17]

A method for producing an antibody comprising an antigen-binding domain whose antigen-binding activity changes according to ion concentration conditions, wherein the method comprises culturing the host cell of claim 16 and collecting the antibody from the cell culture.

[Claim 18]

The method of claim 17, which further comprises any one or more of:

- (a) selecting an antibody which can promote elimination of an antigen from plasma;
- (b) selecting an antibody with enhanced binding activity to an extracellular matrix;
- (c) selecting an antibody with enhanced Fc γ R-binding activity under a neutral pH condition;
- (d) selecting an antibody with enhanced Fc γ RIIb-binding activity under a neutral pH condition;
- (e) selecting an antibody with maintained or enhanced Fc γ RIIb-binding activity and decreased binding activity to one or more activating Fc γ R, selected from the group

consisting of FcγRIa, FcγRIb, FcγRIc, FcγRIIIa, FcγRIIIb and FcγRIIa;

(f) selecting an antibody with enhanced FcRn-binding activity under a neutral pH condition;

(g) selecting an antibody with an increased isoelectric point (pI);

(h) confirming the isoelectric point (pI) of the collected antibody, and then selecting an antibody with an increased isoelectric point (pI); and

(i) selecting an antibody whose antigen-binding activity is changed or increased according to ion concentration conditions;

as compared to a reference antibody.