which a single antigen-binding molecule can bind before its degradation; use of the modified FcRn-binding domain for improving pharmacokinetics of an antigen-binding molecule; and methods for producing said antigen-binding molecules.

Solution to Problem

[0010] The present inventors conducted dedicated studies on modified FcRn-binding domains which have an enhanced affinity for FcRn at neutral pH and on antigen-binding molecules comprising said FcRn-binding domain which have low immunogenicity, high stability and form only few aggregates. As a result, the present inventors discovered that substitutions at specific positions of the FcRn-binding domain increases the affinity for the FcRn at neutral pH without substantially increasing the immunogenicity, without substantially decreasing the stability and/or without substantially increasing the ratio of high molecular weight species.
Furthermore, the present inventors conducted dedicated studies on modified FcRn-binding domains with an enhanced affinity for FcRn at neutral pH or acidic pH but without a significantly increased binding activity for a pre-existing anti-drug antibody and on antigen-binding molecules comprising such an FcRn-binding domain. As a result, the present inventors discovered that substitutions at specific positions of the FcRn-binding domain decrease the affinity for a pre-existing anti-drug antibody at neutral pH without substantially decreasing the FcRn-binding activity.

In a first aspect, there is provided an antigen-binding molecule comprising a modified FcRn-binding domain, wherein the modified FcRn-binding domain comprises an amino acid substitution at one or more positions selected from the group consisting of ~~EU238,~~ EU250, EU252, EU254, ~~EU255,~~ ~~EU258, EU286,~~ EU307, EU308, ~~EU309,~~ EU311, ~~EU315,~~ EU428, ~~EU433,~~ EU434, and EU436, wherein the numbers indicate the position of the substitution according to the EU numbering, wherein the antigen-binding molecule has improved pharmacokinetics compared to a control antibody comprising an intact ~~FcRn~~ FcRn-binding domain, wherein ~~a~~ binding activity of the modified FcRn-binding domain of the antigen-binding molecule for a pre-existing anti-drug antibody (ADA) is not significantly increased as compared to a binding affinity of the intact FcRn-binding domain of the control antibody ~~comprising an intact FcRn-binding domain for ADA~~, wherein the ~~FcRn~~ FcRn-binding domain further comprises an amino acid substitution at one or more positions selected from the group consisting of EU387, EU422, EU424, EU426, ~~EU433,~~ EU436, EU438 and EU440.

In a second aspect, there is provided the use of the antigen-binding molecule as disclosed herein in the manufacture of a medicament for ~~for~~ treating an autoimmune disease, a transplant rejection (graft-vs.-host disease)~~, an inflammatory disease~~ or an allergic disease; wherein the medicament improves antigen-binding molecule-mediated antigen uptake into cells.

In a third aspect, there is provided the use of the antigen-binding molecule as disclosed herein in the manufacture of a medicament for treating an autoimmune disease, a transplant rejection (graft-vs.-host disease)~~, an inflammatory disease~~ or an allergic disease; wherein the medicament reduces the plasma concentration of a specific antigen, wherein the antigen-binding molecule comprises an antigen-binding domain which can bind said antigen.

In a fourth aspect, there is provided a method for improving the pharmacokinetics of an antigen-binding molecule without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of ~~EU238,~~ EU250, EU252, EU254, ~~EU255,~~ ~~EU258, EU286,~~ EU307, EU308, ~~EU309,~~ EU311, ~~EU315,~~ EU428, ~~EU433,~~ EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, ~~EU433,~~ EU436, EU438 and EU440.

In a fifth aspect, there is provided a method for delaying the elimination of an antigen-binding molecule in a subject without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution

into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of ~~EU238,~~ EU250, EU252, EU254, ~~EU255, EU258, EU286,~~ EU307, EU308, ~~EU309,~~ EU311, ~~EU315,~~ EU428, ~~EU433,~~ EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, ~~EU433,~~ EU436, EU438 and EU440.

In a sixth aspect, there is provided a method of prolonging the plasma retention time of an antigen-binding molecule without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of ~~EU238,~~ EU250, EU252, EU254, ~~EU255, EU258, EU286,~~ EU307, EU308, ~~EU309,~~ EU311, ~~EU315,~~ EU428, ~~EU433,~~ EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, ~~EU433,~~ EU436, EU438 and EU440.

In a seventh aspect, there is provided a method for increasing the plasma antigen-elimination rate of an antigen-binding molecule without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of ~~EU238,~~ EU250, EU252, EU254, ~~EU255, EU258, EU286,~~ EU307, EU308, ~~EU309,~~ EU311, ~~EU315,~~ EU428, ~~EU433,~~ EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, ~~EU433,~~ EU436, EU438 and EU440.

In an eighth aspect, there is provided a method for increasing the ability of an antigen-binding molecule to eliminate plasma antigen without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of ~~EU238,~~ EU250, EU252, EU254, ~~EU255, EU258, EU286,~~ EU307, EU308, ~~EU309,~~ EU311, ~~EU315,~~ EU428, ~~EU433,~~ EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, ~~EU433,~~ EU436, EU438 and EU440.

In a ninth aspect, there is provided a method for producing antigen-binding molecules as disclosed herein, which comprises the steps of: (a) selecting a parent FcRn-binding domain and altering the parent FcRn by introducing an amino acid substitution at one or more positions selected from the group consisting of ~~EU238,~~ EU250, EU252, EU254, ~~EU255, EU258, EU286,~~ EU307, EU308, ~~EU309,~~ EU311, ~~EU315,~~ EU428, ~~EU433,~~ EU434, and EU436; (b) selecting an antigen-binding domain of an antigen-binding molecule and altering at least one amino acid in the antigen-binding domain in order to get a pH-dependent antigen-binding domain or a calcium-ion dependent antigen-binding domain; (c) obtaining a gene encoding an antigen-binding molecule in which the human FcRn-binding domain and the antigen-binding domain prepared in (a) and (b) are linked; and (d) producing an antigen-binding molecule using the gene prepared in (c).

In a tenth aspect, there is provided an antigen-binding molecule comprising a modified ~~FcRn~~ FcRn-binding domain, wherein the modified ~~FcRn~~ FcRn-binding domain comprises an amino acid substitution at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, ~~EU433,~~ EU436, EU438 and EU440, wherein the binding affinity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA) at a neutral pH is not significantly increased as compared to the binding affinity of antigen-binding molecule comprising an intact ~~FcRn~~ FcRn-binding domain, wherein the antigen-binding molecule further has an increased binding affinity for an FcRn in the neutral or acidic pH ranges.

In one embodiment, there is provided a method for decreasing the binding activity for a pre-existing ADA of an antibody comprising an FcRn binding domain having an increased binding activity for an FcRn at neutral or acidic pH and an increased binding activity for a pre-existing ADA at a neutral pH, said method comprising the steps of: a) providing an antibody with an FcRn binding domain having an increased binding activity for FcRn at neutral or acidic pH and an increased binding activity for a pre-existing ADA at a neutral

[CLAIMS]

[Claim 1] An antigen-binding molecule comprising a modified FcRn-binding domain, wherein the modified FcRn-binding domain comprises an amino acid substitution at one or more positions selected from the group consisting of EU238, EU250, EU252, EU254, EU255, EU258, EU286, EU307, EU308, EU309, EU311, EU315, EU428, EU433, EU434, and EU436, wherein the numbers indicate the position of the substitution according to the EU numbering,

wherein the antigen-binding molecule has improved pharmacokinetics compared to a control antibody comprising an intact FcRn binding domain,

wherein a binding activity of the antigen-binding molecule for a pre-existing anti-drug antibody (ADA) is not significantly increased as compared to a binding affinity of the control antibody comprising an intact FcRn-binding domain for ADA,

wherein the FcRn binding domain further comprises an amino acid substitution at one or more positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440.

[Claim 2] The antigen-binding molecule according to claim 1, wherein the FcRn-binding domain has
a) an amino acid substitution of the amino acid at position EU252 and EU434; and
b) an amino acid substitution at one or more positions selected from the group consisting of EU238, EU250, EU252, EU254, EU255, EU256, EU258, EU286, EU387, EU307, EU308, EU309, EU311, EU315, EU428, EU433, EU434, and EU436.

[Claim 3] The antigen-binding molecule according to claim 1 or 2, wherein the modified FcRn-binding domain comprises
at position EU238 an aspartic acid,
at position EU250 a valine,
at position EU252 a tyrosine,
at position EU254 a threonine,
at position EU255 a leucine,
at position EU256 a glutamic acid,
at position EU258 an aspartic acid or an isoleucine,
at position EU286 a glutamic acid,
at position EU307 a glutamine,
at position EU308 a proline,
at position EU309 a glutamic acid,
at position EU311 an alanine or a histidine,
at position EU315 an aspartic acid,
at position EU428 an isoleucine,
at position EU433 an alanine, a lysine, a proline, an arginine, or a serine,
at position EU434 a tyrosine, or a tryptophan, and/or
at position EU436 an isoleucine, a leucine, a valine, a threonine, or a phenylalanine.

[Claim 4] The antigen-binding molecule according to claim 2, wherein the FcRn-binding domain comprises an amino acid substitution of an amino acid at one or more position combinations selected from the group consisting of
a) EU252, EU434, and EU436;
b) EU252, EU307, EU311 and EU434;
c) EU252, EU315, and EU434;
d) EU252, EU308, and EU434;
e) EU238, EU252, and EU434;
f) EU252, EU434, EU307, EU311, and EU436; and
g) EU252, EU387, and EU434.

[Claim 5] The antigen-binding molecule according to claim 4, wherein the FcRn-binding domain comprises:
a) at position EU252 a tyrosine, at position EU315 an aspartic acid, and at position EU434 a tyrosine; or
b) at position EU252 a tyrosine, at position EU434 a tyrosine, and at position EU436 an isoleucine; or
c) at position EU252 a tyrosine, at position EU434 a tyrosine, and at position EU436 a leucine; or
d) at position EU252 a tyrosine, at position EU434 a tyrosine, and at position EU436 a valine; or
e) at position EU252 a tyrosine, at position EU254 a threonine, at position EU434 a tyrosine, and at position EU436 an isoleucine.

[Claim 6] The antigen-binding molecule according to claim 2, wherein the FcRn-binding domain comprises an amino acid substitution at three or more positions, wherein the three or more positions are one of the combinations of the group consisting of
a) EU252 / EU434 / EU307 / EU311 / EU286;
b) EU252 / EU434 / EU307 / EU311 / EU286 / EU254;
c) EU252 / EU434 / EU307 / EU311 / EU436;
d) EU252 / EU434 / EU307 / EU311 / EU436 / EU254;
e) EU252 / EU434 / EU307 / EU311 / EU436 / EU250;
f) EU252 / EU434 / EU308 / EU250;
g) EU252 / EU434 / EU308 / EU250 / EU436; and
h) EU252 / EU434 / EU308 / EU250 / EU307 / EU311.

[Claim 7] The antigen-binding molecule according to claim 6, wherein the FcRn-binding domain comprises:
a) at position EU252 a tyrosine, at position EU286 a glutamic acid, at

position EU307 a glutamine, at position EU311 an alanine and at position EU434 a tyrosine; or
b) at position EU252 a tyrosine, at position EU254 a threonine, at position EU286 a glutamic acid, at position EU307 a glutamine, at position EU311 an alanine and at position EU434 a tyrosine; or
c) at position EU252 a tyrosine, at position EU307 a glutamine, at position EU311 an alanine, at position EU434 a tyrosine and at position 436 an isoleucine; or
d) at position EU252 a tyrosine, at position EU254 a threonine, at position EU286 a glutamic acid, at position EU307 a glutamine, at position EU311 an alanine, at position EU434 a tyrosine and at position EU436 an isoleucine; or
e) at position EU250 a valine, at position EU252 a tyrosine, at position EU254 a threonine, at position EU308 a proline, at position EU434 a tyrosine and at position EU436 a valine; or
f) at position EU250 a valine, at position EU252 a tyrosine, at position EU307 a glutamine, at position EU311 an alanine, at position EU434 a tyrosine and at position EU436 a valine; or
g) at position EU252 a tyrosine, at position EU307 a glutamine, at position EU311 an alanine, at position EU434 a tyrosine and at position EU436 a valine; or
h) at position EU250 a valine, at position EU252 a tyrosine, at position EU308 a proline, and at position EU434 a tyrosine; or
i) at position EU250 a valine, at position EU252 a tyrosine, at position EU307 a glutamine, at position EU308 a proline, at position EU311 an alanine, and at position EU434 a tyrosine.

[Claim 8] The antigen-binding molecule according to claim 2, wherein the FcRn-binding domain comprises an amino acid substitution at three or more positions, wherein the three or more positions are one of the combinations of the group consisting of
a) EU252 and EU434 and EU307 and EU311 and EU436 and EU286;
b) EU252 and EU434 and EU307 and EU311 and EU436 and EU250 and EU308;
c) EU252 and EU434 and EU307 and EU311 and EU436 and EU250 and EU286 and EU308;
d) EU252 and EU434 and EU307 and EU311 and EU436 and EU250 and EU286 and EU308 and EU428.

[Claim 9] The antigen-binding molecule according to claim 8, wherein the FcRn-

binding domain comprises:
a) at position EU252 a tyrosine, at position EU286 a glutamic acid, at position EU307 a glutamine, at position EU311 an alanine, at position EU434 a tyrosine, and at position EU436 a valine; or
b) at position EU250 a valine, at position EU252 a tyrosine, at position EU307 a glutamine, at position EU308 proline, at position EU311 an alanine, at position EU434 a tyrosine, and at position EU436 a valine; or
c) at position EU250 a valine, at position EU252 a tyrosine, at position EU286 a glutamic acid, at position EU307 a glutamine, at position EU308 proline, at position EU311 an alanine, at position EU434 a tyrosine, and at position EU436 a valine; or
d) at position EU250 a valine, at position EU252 a tyrosine, at position EU286 a glutamic acid, at position EU307 a glutamine, at position EU308 proline, at position EU311 an alanine, at position EU434 a tyrosine, and at position EU436 a valine.

[Claim 10] The antigen-binding molecule according to claim 2, wherein the FcRn-binding domain comprises an amino acid substitution at three or more positions wherein the three or more positions are one of the combinations of the group consisting of:
a) EU434 and EU307 and EU311;
b) EU434 and EU307 and EU309 and EU311; or
c) EU434 and EU250 and EU252 and EU436.

[Claim 11] The antigen-binding molecule according to claim 10, wherein the FcRn-binding domain comprises:
a) at position EU307 a glutamine, at position EU311 a histidine and at position EU434 a tyrosine; or
b) at position EU307 a glutamine, at position EU309 a glutamic acid, at position EU311 an alanine and at position EU434 a tyrosine; or
c) at position EU307 a glutamine, at position EU309 a glutamic acid, at position EU311 an histidine and at position EU434 a tyrosine; or
d) at position EU250 a valine; at position EU252 a tyrosine, at position EU434 a tyrosine and at position EU436 a valine.

[Claim 12] The antigen-binding molecule according to any one of claims 1 to 11, wherein the ratio of high molecular weight species is less than 2%.

[Claim 13] The antigen-binding molecule according to any one of claims 1 to 12, wherein antigen-binding molecule comprises an antigen-binding domain having

a) a lower binding activity for the antigen at pH 5.5-6.5 than at pH 7-8 or
b) a "calcium concentration-dependent binding" activity for the antigen.

[Claim 14] The antigen-binding molecule according to any one of claims 1 to 5, wherein the binding activity of said binding molecule for the FcRn at pH 7 is 50-150nM, Tm is higher than 63.0 degrees C, and Epibase score is less than 250.

[Claim 15] The antigen-binding molecule according to any one of claims 1 to 3 and claims 6 to 7, and wherein the binding activity of said binding molecule for FcRn at pH 7 is 15-50nM, Tm is higher than 60 degrees C, and Epibase score is less than 500.

[Claim 16] The antigen-binding molecule according to any one of claims 1 to 3 and claims 8 to 9, and wherein the binding activity of said the binding molecule for FcRn at pH 7 is stronger than 15nM, Tm is higher than 57.5 degrees C, and Epibase score is less than 500.

[Claim 17] The antigen-binding molecule according to any one of claims 1 to 3, wherein the FcRn-binding domain comprises an amino acid substitution
a) at the positions EU238, EU255 and/or EU258, and
b) at three or more positions, wherein the three or more positions are one of the combinations set forth in Tables 4 to 7.

[Claim 18] The antigen-binding molecule according to any one of claims 1 to 17, wherein
a) at the position EU257 of the FcRn-binding domain the amino acid is not an amino acid selected from the group consisting of alanine, valine, isoleucine, leucine, and threonine, and/or
b) at the position EU252 of the FcRn-binding domain the amino acid is not tryptophan.

[Claim 19] The antigen-binding molecule according to claim 1, wherein the FcRn

binding domain comprises one or more amino acid substitutions selected from the group consisting of
at position EU387 an arginine,
at position EU422 a glutamic acid, an arginine, or a serine, an aspartic acid, a lysine, a threonine or a glutamine;
at position EU424 a glutamic acid or an arginine, a lysine, or an asparagine;
at position EU426 an aspartic acid, a glutamine, an alanine, or a tyrosine;
at position EU433 an aspartic acid;
at position EU436 a threonine;
at position EU438 a glutamic acid, an arginine, a serine, or a lysine; and
at position EU440 a glutamic acid, aspartic acid or a glutamine.

[Claim 20] The antigen-binding molecule according to any one of claim 1 to 19, wherein the modified FcRn binding domain comprises three or more substitutions, wherein the three or more substitutions are one of the combinations set forth in Tables 12 to 13.

[Claim 21] The antigen-binding molecule according to any one of claims 1 to 20, wherein the modified FcRn-binding domain comprises three or more substitutions, wherein the three or more substitutions are one of the combinations set forth in Tables 14 to 15.

[Claim 22] The antigen-binding molecule according to any one of claims 1 to 21, wherein the FcRn-binding domain comprises:
a) at position EU252 a tyrosine, at position EU387 an arginine, at position EU434 a tyrosine, and at position EU436 a valine; or
b) at position EU252 a tyrosine, at position EU422 a glutamic acid, at position EU434 a tyrosine, and at position EU436 a valine; or
c) at position EU252 a tyrosine, at position EU422 an arginine, at position EU434 a tyrosine, and at position EU436 a valine; or
d) at position EU252 a tyrosine, at position EU422 a serine, at position EU434 a tyrosine, and at position EU436 a valine; or
e) at position EU252 a tyrosine, at position EU424 a glutamic acid, at position EU434 a tyrosine, and at position EU436 a valine; or
f) at position EU252 a tyrosine, at position EU424 an arginine, at position EU434 a tyrosine, and at position EU436 a valine; or
g) at position EU252 a tyrosine, at position EU434 a tyrosine , at position EU436 a valine, and at position EU438 a glutamic acid; or

h) at position EU252 a tyrosine, at position EU434 a tyrosine , at position EU436 a valine, and at position EU438 an arginine; or
i) at position EU252 a tyrosine, at position EU434 a tyrosine, at position EU436 a valine, and at position EU438 a serine; or
j) at position EU252 a tyrosine, at position EU434 a tyrosine, at position EU436 a valine, and at position EU440 a glutamic acid.

[Claim 23] The antigen-binding molecule according to any one of claims 1 to 22, wherein the antigen-binding molecule is an antibody.

[Claim 24] Use of the antigen-binding molecule according to any one of claims 1 to 23 in the manufacture of a medicament for treating an autoimmune disease, a transplant rejection (graft-vs.-host disease), an inflammatory disease or an allergic disease; wherein the medicament improves antigen-binding molecule-mediated antigen uptake into cells.

[Claim 25] Use of the antigen-binding molecule according to any one of claims 1 to 23 in the manufacture of a medicament for treating an autoimmune disease, a transplant rejection (graft-vs.-host disease), an inflammatory disease or an allergic disease; wherein the medicament reduces the plasma concentration of a specific antigen, wherein the antigen-binding molecule comprises an antigen-binding domain which can bind said antigen.

[Claim 26] A method for improving the pharmacokinetics of an antigen-binding molecule without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU238, EU250, EU252, EU254, EU255, EU258, EU286, EU307, EU308, EU309, EU311, EU315, EU428, EU433, EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440.

[Claim 27] A method for delaying the elimination of an antigen-binding molecule in a subject without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of EU238, EU250, EU252, EU254, EU255, EU258, EU286, EU307, EU308, EU309, EU311, EU315, EU428, EU433, EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440.

[Claim 28] A method of prolonging the plasma retention time of an antigen-binding molecule without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of EU238, EU250, EU252, EU254, EU255, EU258, EU286, EU307, EU308, EU309, EU311, EU315, EU428, EU433, EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440.

[Claim 29] A method for increasing the plasma antigen-elimination rate of an antigen-binding molecule without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group

consisting of EU238, EU250, EU252, EU254, EU255, EU258, EU286, EU307, EU308, EU309, EU311, EU315, EU428, EU433, EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440.

[Claim 30] A method for increasing the ability of an antigen-binding molecule to eliminate plasma antigen without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of EU238, EU250, EU252, EU254, EU255, EU258, EU286, EU307, EU308, EU309, EU311, EU315, EU428, EU433, EU434, and EU436; and b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440.

[Claim 31] The method according to any one of claims 26 to 30, wherein further an amino acid substitution at position EU256 into the FcRn binding domain is introduced.

[Claim 32] A method for producing antigen-binding molecules according to any one of claims 1 to 23, which comprises the steps of
(a) selecting a parent FcRn-binding domain and altering the parent FcRn by introducing an amino acid substitution at one or more positions selected from the group consisting of EU238, EU250, EU252, EU254, EU255, EU258, EU286, EU307, EU308, EU309, EU311, EU315, EU428, EU433, EU434, and EU436;
(b) selecting an antigen-binding domain of an antigen-binding molecule and altering at least one amino acid in the antigen-binding domain in order to get a pH-dependent antigen-binding domain or a calcium-ion dependent antigen-binding domain;
(c) obtaining a gene encoding an antigen-binding molecule in which the human FcRn-binding domain and the antigen-binding domain prepared in (a) and (b) are linked and
(d) producing an antigen-binding molecule using the gene prepared in (c).

[Claim 33] The method according to claim 32, wherein in step a) further an amino acid substitution at position EU256 into the FcRn binding domain is introduced.

[Claim 34] The method according to any one of claims 32 to 33, wherein the

method further comprises a step of introducing in the FcRn-binding domain an amino acid substitution at one or more positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440.

[Claim 35] An antigen-binding molecule comprising a modified FcRn binding domain, wherein the modified FcRn binding domain comprises an amino acid substitution at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440, wherein the binding affinity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA) at a neutral pH is not significantly increased as compared to the binding affinity of antigen-binding molecule comprising an intact FcRn binding domain,

wherein the antigen-binding molecule further has an increased binding affinity for an FcRn in the neutral or acidic pH ranges.

[Claim 36] The antigen-binding molecule according to claim 35, wherein the amino acid substituting one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440 is selected from the group consisting of
a) at position EU387 an arginine;
b) at position EU422 a glutamic acid, an arginine, a serine, aspartic acid, lysine, threonine, or glutamine;
c) at position EU424 a glutamic acid, an arginine, a lysine, or asparagines;
d) at position EU426 an aspartic acid, a glutamine, an alanine, or a tyrosine;
e) at position EU433 an aspartic acid
f) at position EU436 a threonine
g) at position EU438 a glutamic acid, an arginine, a serine, or a lysine; and
h) at position EU440 a glutamic acid, an aspartic acid, or a glutamine.

[Claim 37] The antigen-binding molecule according to any one of claims 35 to 36, wherein the modified FcRn binding domain comprises an amino acid substitution at one or more positions or one of the combinations set forth in Table 10.

[Claim 38] The antigen-binding molecule according to any one of claims 35 to 36,

wherein the modified FcRn binding domain comprises any one of the amino acid substitution or substitution combinations set forth in Table 11.

[Claim 39] The antigen-binding molecule according to any one of claims 35 to 38, wherein the modified FcRn binding domain further comprises an amino acid substitution at one or more positions of the FcRn binding domain selected from the group consisting of EU238, EU250, EU252, EU254, EU255, EU256, EU258, EU286, EU307, EU308, EU309, EU311, EU315, EU428, EU434, and EU436, wherein said substitutions confer an increase in FcRn binding activity in the neutral pH or acidic pH range.

[Claim 40] The antigen-binding molecule according to any one of claims 35 to 39, wherein the modified FcRn binding domain comprises amino acid substitutions at the FcRn binding domain positions
i) a) EU438 / EU440 or b) EU424; and
ii) a) EU434, b) EU252/EU254/EU256; c) EU428/EU434; or d) EU250/EU428.

[Claim 41] The antigen-binding molecule according to claim 40, wherein the modified FcRn binding domain comprises amino acid substitutions
i) a) EU438R/EU440E or b) EU424N; and
ii) a) M434H; b) M252Y/S254T/T256E; c) M428L/N434S; or d) T250Q and M428L (EU numbering).

[Claim 42] The antigen-binding molecule according to claim 41, wherein the modified FcRn binding domain comprises three or more amino acid substitutions wherein the three or more substitutions are one of the combinations set forth in Tables 13 and 15.

[Claim 43] The antigen-binding molecule according to any one of claims 35 to 38 wherein the modified FcRn binding domain comprises substitutions
a) at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU438, EU440, EU433, or at two or more positions wherein the two positions are one of the combinations of the group consisting of EU422/EU424, and EU438/EU440; and
b) two or more positions wherein the two positions are one of the combinations set forth in Table 9.

[Claim 44] The antigen-binding molecule according to claim 43, wherein the modified FcRn binding domain comprises three or more the amino acid substitutions wherein the three or more the amino acid substitutions are one of the combinations set forth in Tables 12 or 14.

[Claim 45] The antigen-binding molecule of any one of claims 35 to 44 wherein

said antigen-binding molecule comprises a pH-dependent antigen-binding domain or a calcium ion-dependent antigen-binding domain.

[CLAIMS]

[Claim 1] An antigen-binding molecule comprising a modified FcRn-binding domain, wherein the modified FcRn-binding domain comprises an amino acid substitution at one or more positions selected from the group consisting of EU250, EU252, EU254, EU307, EU308, EU311, EU428, EU434, and EU436, wherein the numbers indicate the position of the substitution according to the EU numbering,

wherein the antigen-binding molecule has improved pharmacokinetics compared to a control antibody comprising an intact FcRn-binding domain,

wherein binding activity of the modified FcRn-binding domain of the antigen-binding molecule for a pre-existing anti-drug antibody (ADA) is not significantly increased as compared to a binding affinity of the intact FcRn-binding domain of the control antibody,

wherein the FcRn-binding domain further comprises an amino acid substitution at one or more positions selected from the group consisting of EU387, EU422, EU424, EU426, EU436, EU438 and EU440.

[Claim 2] The antigen-binding molecule according to claim 1, wherein the modified FcRn-binding domain comprises an amino acid substitution of an amino acid at one or more position combinations selected from the group consisting of

a) EU252, EU254 and EU256;
b) EU252, EU434 and EU436;
c) EU250, EU252, EU307, EU308, EU311, EU434 and EU436;
d) EU428 and EU434; and
e) EU424,

and wherein the FcRn-binding domain further comprises an amino acid substitution of an amino acid at one or more position combinations selected from the group consisting of

a) EU387;
b) EU422;
c) EU424;
d) EU426;
e) EU436;
f) EU438;
g) EU440;
h) EU422 and EU424; and
i) EU438 and EU440.

[Claim 3] The antigen-binding molecule according to claim 1 or 2, wherein the modified FcRn-binding domain comprises

at position EU250 a valine,
at position EU252 a tyrosine,
at position EU254 a threonine,
at position EU307 a glutamine,
at position EU308 a proline,
at position EU311 an alanine or a histidine,
at position EU428 an isoleucine,
at position EU434 a tyrosine, or a tryptophan, and/or
at position EU436 an isoleucine, a leucine, a valine, a threonine, or a phenylalanine.

[Claim 4] The antigen-binding molecule according to any one of claims 1 to 3, wherein the ratio of high molecular weight species is less than 2%.

[Claim 5] The antigen-binding molecule according to any one of claims 1 to 4, wherein antigen-binding molecule comprises an antigen-binding domain having
a) a lower binding activity for the antigen at pH 5.5-6.5 than at pH 7-8 or
b) a "calcium concentration-dependent binding" activity for the antigen.

[Claim 6] The antigen-binding molecule according to any one of claims 1 to 3, wherein the binding activity of said binding molecule for the FcRn at pH 7 is 50-150nM, Tm is higher than 63.0 degrees C, and Epibase score is less than 250.

[Claim 7] The antigen-binding molecule according to any one of claims 1 to 3, and wherein the binding activity of said binding molecule for FcRn at pH 7 is 15-50nM, Tm is higher than 60 degrees C, and Epibase score is less than 500.

[Claim 8] The antigen-binding molecule according to any one of claims 1 to 3, and wherein the binding activity of said the binding molecule for FcRn at pH 7 is stronger than 15nM, Tm is higher than 57.5 degrees C, and Epibase score is less than 500.

[Claim 9] The antigen-binding molecule according to claim 1, wherein the FcRn-binding domain comprises one or more amino acid substitutions selected from the group consisting of
at position EU387 an arginine;
at position EU422 a glutamic acid, an arginine, or a serine, an aspartic acid, a lysine, a threonine or a glutamine;
at position EU424 a glutamic acid or an arginine, a lysine, or an asparagine;
at position EU426 an aspartic acid, a glutamine, an alanine, or a tyrosine;
at position EU436 a threonine;

at position EU438 a glutamic acid, an arginine, a serine, or a lysine; and at position EU440 a glutamic acid, aspartic acid or a glutamine.

[Claim 10] The antigen-binding molecule according to any one of claim 1 to 9, wherein the modified FcRn-binding domain comprises three or more substitutions, wherein the three or more substitutions are one of the combinations set forth in Tables 12 to 13.

[Claim 11] The antigen-binding molecule according to any one of claims 1 to 10, wherein the modified FcRn-binding domain comprises three or more substitutions, wherein the three or more substitutions are one of the combinations set forth in Tables 14 to 15.

[Claim 12] The antigen-binding molecule according to any one of claims 1 to 11, wherein the FcRn-binding domain comprises:
a) at position EU252 a tyrosine, at position EU387 an arginine, at position EU434 a tyrosine, and at position EU436 a valine; or
b) at position EU252 a tyrosine, at position EU422 a glutamic acid, at position EU434 a tyrosine, and at position EU436 a valine; or
c) at position EU252 a tyrosine, at position EU422 an arginine, at position EU434 a tyrosine, and at position EU436 a valine; or
d) at position EU252 a tyrosine, at position EU422 a serine, at position EU434 a tyrosine, and at position EU436 a valine; or
e) at position EU252 a tyrosine, at position EU424 a glutamic acid, at position EU434 a tyrosine, and at position EU436 a valine; or
f) at position EU252 a tyrosine, at position EU424 an arginine, at position EU434 a tyrosine, and at position EU436 a valine; or
g) at position EU252 a tyrosine, at position EU434 a tyrosine , at position EU436 a valine, and at position EU438 a glutamic acid; or
h) at position EU252 a tyrosine, at position EU434 a tyrosine, at position EU436 a valine, and at position EU438 an arginine; or
i) at position EU252 a tyrosine, at position EU434 a tyrosine, at position EU436 a valine, and at position EU438 a serine; or
j) at position EU252 a tyrosine, at position EU434 a tyrosine, at position EU436 a valine, and at position EU440 a glutamic acid.

[Claim 13] The antigen-binding molecule according to any one of claims 1 to 12, wherein the antigen-binding molecule is an antibody.

[Claim 14] Use of the antigen-binding molecule according to any one of claims 1 to 13 in the manufacture of a medicament for treating an autoimmune disease, a transplant rejection (graft-vs.-host disease) or an allergic disease; wherein the medicament improves antigen-binding molecule-mediated antigen uptake into cells.

[Claim 15] Use of the antigen-binding molecule according to any one of claims 1 to 13 in the manufacture of a medicament for treating an autoimmune disease, a

transplant rejection (graft-vs.-host disease) or an allergic disease; wherein the medicament reduces the plasma concentration of a specific antigen, wherein the antigen-binding molecule comprises an antigen-binding domain which can bind said antigen.

[Claim16] A method for improving the pharmacokinetics of an antigen-binding molecule without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of
a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU250, EU252, EU254, EU307, EU308, EU311, EU428, EU434, and EU436; and
b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU436, EU438 and EU440.

[Claim 17] A method for delaying the elimination of an antigen-binding molecule in a subject without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of

a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of EU250, EU252, EU254, EU307, EU308, EU311, EU428, EU434, and EU436; and

b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU436, EU438 and EU440.

[Claim 18] A method of prolonging the plasma retention time of an antigen-binding molecule without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of

a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of EU250, EU252, EU254, EU307, EU308, EU311, EU428, EU434, and EU436; and

b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU436, EU438 and EU440.

[Claim 19] A method for increasing the plasma antigen-elimination rate of an antigen-binding molecule without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of

a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of EU250, EU252, EU254, EU307, EU308, EU311, EU428, EU434, and EU436; and

b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU436, EU438 and EU440.

[Claim 20] A method for increasing the ability of an antigen-binding molecule to eliminate plasma antigen without significantly increasing the binding activity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA), comprising the steps of

a) introducing an amino acid substitution into an FcRn-binding domain of said antigen-binding molecule at one or more of the positions selected from the group consisting of EU250, EU252, EU254, EU307, EU308, EU311, EU428, EU434, and EU436; and

b) introducing further amino acid substitution into an FcRn-binding domain of said antigen-binding molecules at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU436, EU438 and EU440.

[Claim 21] The method according to any one of claims 16 to 20, wherein further an amino acid substitution at position EU256 into the FcRn-binding domain is introduced.

[Claim 22] A method for producing antigen-binding molecules according to any one of claims 1 to 13, which comprises the steps of
(a) selecting a parent FcRn-binding domain and altering the parent FcRn by introducing an amino acid substitution at one or more positions selected from the group consisting of EU250, EU252, EU254, EU307, EU308, EU311, EU428, EU434, and EU436;
(b) selecting an antigen-binding domain of an antigen-binding molecule and altering at least one amino acid in the antigen-binding domain in order to get a pH-dependent antigen-binding domain or a calcium-ion dependent antigen-binding domain;
(c) obtaining a gene encoding an antigen-binding molecule in which the human FcRn-binding domain and the antigen-binding domain prepared in (a) and (b) are linked and
(d) producing an antigen-binding molecule using the gene prepared in (c).

[Claim 23] The method according to claim 22, wherein in step a) further an amino acid substitution at position EU256 into the FcRn-binding domain is introduced.

[Claim 24] The method according to any one of claims 22 to 23, wherein the method further comprises a step of introducing in the FcRn-binding domain an amino acid substitution at one or more positions selected from the group consisting of EU387, EU422, EU424, EU426, EU436, EU438 and EU440.

[Claim 25] An antigen-binding molecule comprising a modified FcRn-binding domain, wherein the modified FcRn-binding domain comprises an amino acid substitution at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU436, EU438 and EU440, wherein the binding affinity of said antigen-binding molecule for a pre-existing anti-drug antibody (ADA) at a neutral pH is not significantly increased as compared to the binding affinity of antigen-binding molecule comprising an intact FcRn-binding domain,

wherein the antigen-binding molecule further has an increased binding affinity for an FcRn in the neutral or acidic pH ranges.

[Claim 26] The antigen-binding molecule according to claim 25, wherein the modified FcRn-binding domain comprises an amino acid substitution of an amino acid at one or more position combinations selected from the group consisting of

a) EU387;
b) EU422;
c) EU424;
d) EU426;
e) EU436;
f) EU438;
g) EU440;
h) EU422 and EU424; and
i) EU438 and EU440.

[Claim 27] The antigen-binding molecule according to claims 25 or 26, wherein the amino acid substituting one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU426, EU433, EU436, EU438 and EU440 is selected from the group consisting of
a) at position EU387 an arginine;
b) at position EU422 a glutamic acid, an arginine, a serine, aspartic acid, lysine, threonine, or glutamine;
c) at position EU424 a glutamic acid, an arginine, a lysine, or asparagines;
d) at position EU426 an aspartic acid, a glutamine, an alanine, or a tyrosine;

e) at position EU433 an aspartic acid
f) at position EU436 a threonine
g) at position EU438 a glutamic acid, an arginine, a serine, or a lysine; and
h) at position EU440 a glutamic acid, an aspartic acid, or a glutamine.

[Claim 28] The antigen-binding molecule according to any one of claims 25 to 27, wherein the modified FcRn-binding domain further comprises an amino acid substitution at one or more positions of the FcRn-binding domain selected from the group consisting of EU250, EU252, EU254, EU307, EU308, EU309, EU311, EU428, EU434, and EU436, wherein said substitutions confer an increase in FcRn-binding activity in the neutral pH or acidic pH range.

[Claim 29] The antigen-binding molecule according to any one of claims 25 to 28, wherein the modified FcRn-binding domain comprises amino acid substitutions at the FcRn-binding domain positions
i) a) EU438/EU440 or b) EU424; and
ii) a) EU434, b) EU252/EU254/EU256; c) EU428/EU434; or d) EU250/EU428.

[Claim 30] The antigen-binding molecule according to claim 29, wherein the modified FcRn-binding domain comprises amino acid substitutions
i) a) EU438R/EU440E or b) EU424N; and
ii) a) M434H; b) M252Y/S254T/T256E; c) M428L/N434S; or d) T250Q and M428L (EU numbering).

[Claim 31] The antigen-binding molecule according to claim 30, wherein the modified FcRn-binding domain comprises three or more amino acid substitutions wherein the three or more substitutions are one of the combinations set forth in Tables 13 and 15.

[Claim 32] The antigen-binding molecule according to any one of claims 25 to 27 wherein the modified FcRn-binding domain comprises substitutions
a) at one or more of the positions selected from the group consisting of EU387, EU422, EU424, EU438, EU440, EU433, or at two or more positions wherein the two positions are one of the combinations of the group consisting of EU422/EU424, and EU438/EU440; and
b) two or more positions wherein the two positions are one of the combinations set forth in Table 9.

[Claim 33] The antigen-binding molecule according to claim 32, wherein the modified FcRn-binding domain comprises three or more the amino acid substitutions wherein the three or more the amino acid substitutions are one of the combinations set forth in Tables 12 or 14.

[Claim 34] The antigen-binding molecule of any one of claims 25 to 33 wherein said antigen-binding molecule comprises a pH-dependent antigen-binding domain or a calcium ion-dependent antigen-binding domain.