[0011] Provided are antigen-binding molecules comprising: a first conjugated immunoglobulin comprising a first heavy chain variable region and a first light chain variable region, the first light chain variable region having a cysteine at position 80 ("$Cys80^1$"), wherein $Cys80^1$ is conjugated to a first thiol-reactive compound comprising a first thiol-reactive group, and a second conjugated immunoglobulin comprising a second heavy chain variable region and a second light chain variable region, the second light chain variable region having a cysteine at position 80 ("$Cys80^2$") wherein $Cys80^2$ is conjugated to a second thiol-reactive compound comprising a second thiol-reactive group. In one aspect, the present disclosure refers to a chimeric or humanized rabbit immunoglobulin comprising a heavy chain variable region and a light chain variable region, the light chain variable region having a cysteine at position 80 ("Cys80") and phenylalanine at position 83 is substituted with an amino acid other than Phe, Lys, or Cys, wherein the Cys80 is unpaired and the numbering of the Cys80 and position 83 are based upon the Kabat or Chothia numbering system, and wherein the immunoglobulin retains an antigen-binding affinity comparable to that of a parental non-chimeric or non-humanized rabbit immunoglobulin and exhibits 25% or less aggregation and 70% or higher conjugation efficiency to the Cys80.

[0012] Light chain variable regions for use in a conjugated immunoglobulin, the light chain variable region having a cysteine at amino acid position 80 ("Cys80") and an amino acid residue other than Phe, Lys, or Cys at amino acid position 83, wherein the Cys80 is unpaired are also disclosed herein.

BRIEF DESCRIPTION OF THE DRAWINGS

[0013] The summary, as well as the following detailed description, is further understood when read in conjunction with the appended drawings. For the purpose of illustrating the disclosed methods, conjugated immunoglobulins, antigen-binding molecules, immunoglobulins, and light chain variable regions, there are shown in the drawings exemplary embodiments; however, the methods, conjugated immunoglobulins, antigen-binding molecules, immunoglobulins, and light chain variable regions are not limited to the specific embodiments disclosed. In the drawings:

[0014] **FIG. 1** represents an alignment of rabbit and human light chain sequences. **(A)** Alignment of the germline Vκ sequences from rabbit (IGKV1S2, X02336) and human (IGKV1-5, Z00001). The arrow indicates Cys80 (according to either Kabat or Chothia numbering) in the rabbit sequence. **(B)** Alignment of the germline Cκ sequences from rabbit (IGKC1, K01360)

and human (IGKC, J00241). The arrow indicates Cys171 (EU numbering) in the rabbit sequence.

[0015] **FIG. 2** represents structural models of **(A)** rabbit mAb, showing the Cys80-Cys171 disulfide bond, **(B)** human mAb, and **(C)** rabbit-human chimeric mAb showing the unpaired Cys80.

[0016] **FIG. 3** illustrates an alignment of rabbit germline Vκ families. The residue at position 80 is indicated by the arrow.

What is Claimed:

1. A chimeric or humanized rabbit immunoglobulin comprising:

   a heavy chain variable region and a light chain variable region, the light chain variable region having a cysteine at position 80 ("Cys80") and phenylalanine at position 83 is substituted with an amino acid other than Phe, Lys, or Cys, wherein the Cys80 is unpaired and the numbering of the Cys80 and position 83 are based upon the Kabat or Chothia numbering system, and

   wherein the immunoglobulin retains an antigen-binding affinity comparable to that of a parental non-chimeric or non-humanized rabbit immunoglobulin and exhibits 25% or less aggregation and 70% or higher conjugation efficiency to the Cys80.

2. The immunoglobulin of claim 1, wherein the Cys80 is decapped.

3. The immunoglobulin of claim 1, further comprising a thiol-reactive compound conjugated to the cysteine at position 80, wherein the thiol-reactive compound comprises a thiol-reactive group.

4. The immunoglobulin of claim 1, wherein the immunoglobulin exhibits 5% or less aggregation.

5. The immunoglobulin of claim 1, wherein the immunoglobulin exhibits 10% or less aggregation.

6. The immunoglobulin of claim 1, wherein the immunoglobulin exhibits 85% or higher conjugation efficiency to the Cys80.

7. The immunoglobulin of claim 1, wherein the immunoglobulin exhibits 91% or higher conjugation efficiency to the Cys80.