

Amended Claim Set

1. (Currently Amended) A method of purifying or enriching a biomolecule, ~~the said~~ method comprising the steps:

- (a) providing an aqueous solution containing ~~the said~~ biomolecule;
- (b) contacting ~~the said~~ aqueous solution with a multimodal resin;
- (c) optionally washing the multimodal resin with an aqueous washing buffer;
- (d) eluting the biomolecule from the multimodal resin by an aqueous buffer solution,

wherein ~~the said~~ aqueous buffer solution comprises ~~one or more of~~:

(i) at least 15 mM of N-(2-acetoamido)iminodiacetic acid (ADA), N-(2-acetamido)-2-aminoethanesulfonic acid (ACES), N,N-bis(2-hydroxyethyl)-2-aminoethanesulfonic acid (BES), N,N-bis(2-hydroxyethyl)glycine (Bicine), 3-(cyclohexylamino)-1-propanesulfonic acid (CAPS), 3-(cyclohexylamino)-2-hydroxy-1-propanesulfonic acid (CAPSO), 2-(cyclohexylamino)ethanesulfonic acid (CHES), 4-(2-hydroxyethyl)piperazine-1-ethanesulfonic acid (HEPES), 3-(N-morpholino)propanesulfonic acid (MOPS), 1,4-piperazinediethanesulfonic acid (PIPES), N-[tris(hydroxymethyl)methyl]-3-aminopropanesulfonic acid (TAPS), or 2-[(2-hydroxy-1,1-bis(hydroxymethyl)ethyl)amino]ethanesulfonic acid (TES)] ~~a Good's buffer; and~~

(ii) one or both of:

~~(ii)~~ between 100 and 400 mM alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, or valine; and

~~(iii)~~ between 100 and 400 mM guanidium chloride, guanidium thiocyanate, ammonium chloride, ammonium sulfate, ammonium phosphate, ammonium acetate, sodium chloride, sodium citrate, sodium caprylate, sodium phosphate, sodium iodide, potassium chloride, potassium citrate, potassium sulfate, potassium phosphate, potassium iodide, magnesium chloride, magnesium sulfate, magnesium phosphate, calcium chloride, calcium sulfate, calcium phosphate, or taurine; and

optionally, further comprising (iii) ~~(iv)~~ between 1% and 60% (v/v) of ethylene glycol or propylene glycol or a mixture of ethylene glycol and propylene glycol; and

(e) collecting biomolecule containing fractions, thereby obtaining the biomolecule in purified or enriched form.

2. (Currently Amended) The method according to claim 1, wherein the aqueous buffer solution comprises 15 mM to 2.5 M of ADA, ACES, BES, Bicine, CAPS, CAPSO, CHES, HEPES, MOPS, PIPES, TAPS, or TES ~~Good's buffer~~.

~~3. The method according to claim 1 or claim 2, wherein the Good's buffer is MES, MOPS, or Tris.~~

~~3.4.~~ (Currently Amended) The method according to claim 1 or claim 2, wherein the aqueous buffer solution comprises ~~Good's buffer~~ is MES.

~~4.5.~~ (Currently Amended) The method according to claim 1 or claim 2, wherein the aqueous buffer solution comprises ~~Good's buffer~~ is MOPS.

~~5.6.~~ (Currently Amended) The method according to any one of claims 1 to ~~45~~, wherein the biomolecule is a protein.

~~6.7.~~ (Currently Amended) The method according to claim ~~56~~, wherein the protein is an enzyme.

~~7.8.~~ (Currently Amended) The method according to claim ~~67~~, wherein the enzyme is glycosidase.

~~8.9.~~ (Currently Amended) The method according to claim ~~78~~, wherein the glycosidase is an alpha-galactosidase.

~~9.10.~~ (Currently Amended) The method according to claim ~~89~~, wherein the alpha-galactosidase is selected from the group consisting of agalsidase ~~alfa~~ ~~alpha~~ and agalsidase beta.

~~10.11.~~ (Currently Amended) The method according to any one of claims 1 to ~~910~~, wherein the multimodal resin is selected from the group consisting of Capto™ Adhere, Capto™

MMC, Pall PPA HyperCel™, Pall HEA HyperCel™, Pall MEP HyperCel™, and Eshmuno® HCX.

~~11.12.~~ (Currently Amended) A purified biomolecule obtained by the method according to any one of claims 1 to ~~1011~~.

~~12.13.~~ (Currently Amended) A preparation comprising:

(a) a biomolecule; and

(b) an aqueous buffer solution comprising ~~one or more of~~:

(i) at least 15 mM ADA, ACES, BES, Bicine, CAPS, CAPSO, CHES, HEPES, MOPS, PIPES, TAPS, or TES ~~Good's buffer~~;

(ii) one or both of:

(~~ii~~) between 100 and 400 mM alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, or valine; and

(~~iii~~) between 100 and 400 mM guanidium chloride, guanidium thiocyanate, ammonium chloride, ammonium sulfate, ammonium phosphate, ammonium acetate, sodium chloride, sodium citrate, sodium caprylate, sodium phosphate, sodium iodide, potassium chloride, potassium citrate, potassium sulfate, potassium phosphate, potassium iodide, magnesium chloride, magnesium sulfate, magnesium phosphate, calcium chloride, calcium sulfate, calcium phosphate, or taurine; and

optionally, further comprising (~~iii~~)(iv) between 1% and 60% (v/v) of ethylene glycol or propylene glycol or a mixture of ethylene glycol and propylene glycol.

~~13.14.~~ (Currently Amended) The preparation according to claim ~~1213~~, wherein the aqueous buffer solution comprises 15 mM and 2.5 M ADA, ACES, BES, Bicine, CAPS, CAPSO, CHES, HEPES, MOPS, PIPES, TAPS, or TES ~~Good's buffer~~.

~~15. The preparation according to claim 13 or 14, wherein the at least one Good's buffer is selected from the group consisting of MES, MOPS, or Tris.~~

~~14.16.~~ (Currently Amended) The preparation according to claim ~~12 or 13~~~~13 or 14~~, wherein the aqueous buffer solution comprises ~~Good's buffer~~ is MES.

~~15.17.~~ (Currently Amended) The preparation according to claim ~~12 or 13~~~~13 or 14~~, wherein the aqueous buffer solution comprises ~~Good's buffer~~ is MOPS.

~~16.18.~~ (Currently Amended) The preparation according to any one of claims ~~12 to 15~~~~13 to 17~~, wherein the biomolecule is a protein.

~~17.19.~~ (Currently Amended) The preparation according to claim ~~16~~~~18~~, wherein the protein is an enzyme.

~~18.20.~~ (Currently Amended) The preparation according to claim ~~17~~~~19~~, wherein the enzyme is a glycosidase.

~~19.21.~~ (Currently Amended) The preparation according to claim ~~18~~~~20~~, wherein the glycosidase is an alpha-galactosidase.

~~20.22.~~ (Currently Amended) The preparation according to claim ~~19~~~~21~~, wherein the alpha-galactosidase is selected from the group of agalsidase ~~alfa~~ ~~alpha~~ and agalsidase beta.

~~21.23.~~ (Currently Amended) A method for desorbing a biomolecule from a multimodal resin, comprising contacting a biomolecule bound to a multimodal resin with an aqueous buffer, wherein the said ~~aqueous buffer solution comprises one or more of:~~

(i) at least 15 mM ADA, ACES, BES, Bicine, CAPS, CAPSO, CHES, HEPES, MOPS, PIPES, TAPS, or TES ~~Good's buffer~~; and

(ii) one or both of:

~~(iii)~~ between 100 and 400 mM alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, or valine; and

~~(iii)~~ between 100 and 400 mM guanidium chloride, guanidium thiocyanate, ammonium chloride, ammonium sulfate, ammonium phosphate, ammonium acetate, sodium chloride, sodium citrate, sodium caprylate, sodium phosphate, sodium iodide, potassium chloride, potassium citrate, potassium sulfate, potassium phosphate, potassium iodide, magnesium chloride, magnesium sulfate, magnesium phosphate, calcium chloride, calcium sulfate, calcium phosphate, or taurine; and

optionally, further comprising (iii) ~~(iv)~~ between 1% and 60% (v/v) of ethylene glycol or propylene glycol or a mixture of ethylene glycol and propylene glycol.

~~22.24.~~ (Currently Amended) A method for increasing the recovery rate of a biomolecule absorbed on a multimodal resin by contacting the said multimodal resin onto which the said biomolecule has been absorbed with an aqueous buffer solution, wherein the said aqueous buffer solution comprises ~~one or more of~~:

(i) at least 15 mM ADA, ACES, BES, Bicine, CAPS, CAPSO, CHES, HEPES, MOPS, PIPES, TAPS, or TES ~~Good's buffer~~; and

(ii) one or both of:

~~(ii)~~ between 100 and 400 mM alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, or valine; and

~~(iii)~~ between 100 and 400 mM guanidium chloride, guanidium thiocyanate, ammonium chloride, ammonium sulfate, ammonium phosphate, ammonium acetate, sodium chloride, sodium citrate, sodium caprylate, sodium phosphate, sodium iodide, potassium chloride, potassium citrate, potassium sulfate, potassium phosphate, potassium iodide, magnesium chloride, magnesium sulfate, magnesium phosphate, calcium chloride, calcium sulfate, calcium phosphate, or taurine; and

optionally, further comprising (iii) ~~(iv)~~ between 1% and 60% (v/v) of ethylene glycol or propylene glycol or a mixture of ethylene glycol and propylene glycol.

~~23.25.~~ (Currently Amended) The method of claim 2123 or claim 2224, wherein the aqueous buffer solution comprises between 15 mM and 2.5 M ADA, ACES, BES, Bicine, CAPS, CAPSO, CHES, HEPES, MOPS, PIPES, TAPS, or TES ~~Good's buffer~~.

~~26.~~ The method of any one of claims ~~23 to 25~~, wherein the Good's buffer is selected from the group consisting of MES, MOPS, and Tris.

~~24.~~27. (Currently Amended) The method of any one of claims ~~21 to 23~~23 to 25, wherein the aqueous buffer solution comprises Good's buffer is MES.

~~25.~~28. (Currently Amended) The method of any one of claims ~~21 to 23~~23 to 25, wherein the aqueous buffer solution comprises Good's buffer is MOPS.

~~26.~~29. (Currently Amended) The method of any one of claims ~~21 to 25~~23 to 28, wherein the biomolecule is a protein.

~~27.~~30. (Currently Amended) The method of claim ~~26~~29, wherein the protein is an enzyme.

~~28.~~31. (Currently Amended) The method of claim ~~27~~30, wherein the enzyme is a glycosidase.

~~29.~~32. (Currently Amended) The method of claim ~~28~~31, wherein the glycosidase is an alpha-galactosidase.

~~30.~~33. (Currently Amended) The method of claim ~~29~~32, wherein the alpha-galactosidase is selected from the group consisting of agalsidase ~~alfa~~ alpha and agalsidase beta.

~~31.~~34. (Currently Amended) The method of any one of claims ~~21 to 30~~23 to 33, wherein the multimodal resin is selected from the group consisting of CaptoTM Adhere, CaptoTM MMC, Pall PPA HyperCelTM, Pall HEA HyperCelTM, Pall MEP HyperCelTM, and Eshmuno® HCX.

~~32.~~35. (Currently Amended) An article of manufacture comprising:

(a) a packaging material or container;

- (b) an aqueous buffer solution comprising ~~one or more of~~:
- (i) at least 15 mM ADA, ACES, BES, Bicine, CAPS, CAPSO, CHES, HEPES, MOPS, PIPES, TAPS, or TES ~~Good's buffer~~; and
 - (ii) one or both of:
 - ~~(ii)~~ between 100 and 400 mM alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, or valine; and
 - ~~(iii)~~ between 100 and 400 mM ~~in total concentration of~~ urea, guanidium chloride, guanidium thiocyanate, ammonium chloride, ammonium sulfate, ammonium phosphate, ammonium acetate, sodium chloride, potassium chloride, potassium citrate, potassium sulfate, potassium phosphate, potassium iodide, magnesium chloride, magnesium sulfate, magnesium phosphate, calcium chloride, calcium sulfate, calcium phosphate, or taurine; and
 - optionally, further comprising (iii) ~~(iv)~~ between 1% and 60% (v/v) of ethylene glycol or propylene glycol or a mixture of ethylene glycol and propylene glycol; and - (c) a data carrier containing instructions for carrying out a method according to any one of claims 1 to 10~~11~~.

~~33.36.~~ (Currently Amended) The article of manufacture according to claim 32~~35~~, wherein the data carrier is a label present on the packaging material or container or a packaging insert.

~~34.37.~~ (Currently Amended) The article of manufacture according to claim 32~~35~~ or claim 33~~36~~, wherein the data carrier comprises written information, visual information, or computer-readable information.

~~35.38.~~ (Currently Amended) The article of manufacture according to any one of claims 32 to 34~~35 to 37~~, wherein the data carrier further comprises one or more of the following information:

- (i) instructions for use of aqueous buffer solution;
- (ii) quality information such as information about the lot/batch number, the manufacturing or assembly site or the expiry or sell-by date of the aqueous buffer solution; or information concerning the correct storage or handling of the article;
- (iii) information concerning the composition of the aqueous buffer solution;

(iv) information concerning the interpretation of information obtained when performing methods according to any one of claims 1 to ~~10~~11;

(v) a warning concerning possible misinterpretations or wrong results when applying unsuitable methods; and

(vi) a warning concerning possible misinterpretations or wrong results when using unsuitable reagent(s) and/or buffer(s).

~~36.39.~~ (Currently Amended) The article of manufacture according to any one of claims ~~32 to 35~~32 to 38, further comprising one or more of the following components:

(d) a chromatography medium;

(e) a chromatography column; and

(f) a reference biomolecule usable for calibrating a method for purifying or enriching a biomolecule.

~~37.40.~~ (Currently Amended) The article of manufacture according to claim ~~36~~39, wherein ~~the said~~ chromatography column is prepacked with ~~the said~~ chromatography medium.

~~38.41.~~ (Currently Amended) The article of manufacture according to claim 36 or claim 37~~any one of claims 39 to 40~~, wherein ~~the said~~ chromatography medium is a multimodal chromatography resin.

~~39.42.~~ (Currently Amended) The article of manufacture according to claim ~~38~~41, wherein the multimodal chromatography resin is selected from the group consisting of CaptoTM Adhere, CaptoTM MMC, Pall PPA HyperCelTM, Pall HEA HyperCelTM, Pall MEP HyperCelTM, and Eshmuno® HCX.

~~40.43.~~ (Currently Amended) The article of manufacture according to any one of claims ~~36 to 39~~36 to 42, wherein the reference biomolecule is a protein or a nucleic acid.

~~41.44.~~ (Currently Amended) The article of manufacture according to any one of claims ~~32 to 40~~32 to 43, wherein the aqueous buffer solution comprises between 15 mM and 2.5 M

ADA, ACES, BES, Bicine, CAPS, CAPSO, CHES, HEPES, MOPS, PIPES, TAPS, or TES
Good's buffer.

~~45. The article of manufacture according to any one of claims 35 to 44, wherein the at least one Good's buffer is selected from the group consisting of MES, MOPS, and Tris.~~

~~42.~~46. (Currently Amended) The article of manufacture according to any one of claims 32 to 41~~35 to 45~~, wherein the aqueous buffer solution comprises Good's buffer is MES.

~~43.~~47. (Currently Amended) The article of manufacture according to any one of claims 32 to 42~~35 to 45~~, wherein the aqueous buffer solution comprises Good's buffer is MOPS.

44. (New) The method according to claim 3 or claim 4, wherein the aqueous buffer solution comprises 10 mM to 400 mM arginine.

45. (New) The method according to claim 44, wherein the aqueous buffer solution comprises 150 mM to 400 mM arginine.

46. (New) The preparation according to claim 14 or claim 15, wherein the aqueous buffer solution comprises 10 mM to 400 mM arginine.

47. (New) The preparation according to claim 46, wherein the aqueous buffer solution comprises 150 mM to 400 mM arginine.

48. (New) The method according to claim 24 or claim 25, wherein the aqueous buffer solution comprises 10 mM to 400 mM arginine.

49. (New) The method according to claim 48, wherein the aqueous buffer solution comprises 150 mM to 400 mM arginine.

50. (New) The article of manufacture according to claim 42 or claim 43, wherein the aqueous buffer solution comprises 10 mM to 400 mM arginine.

51. (New) The article of manufacture according to claim 50, wherein the aqueous buffer solution comprises 150 mM to 400 mM arginine.

52. (New) The method according to claim 9, wherein the agalsidase beta is Fabrazyme[®].

53. (New) The method according to claim 9, wherein the agalsidase alfa is Replagal[™].

54. (New) The preparation according to claim 20, wherein the agalsidase beta is Fabrazyme[®].

55. (New) The preparation according to claim 20, wherein the agalsidase alfa is Replagal[™].

56. (New) The method according to claim 30, wherein the agalsidase beta is Fabrazyme[®].

57. (New) The method according to claim 30, wherein the agalsidase alfa is Replagal[™].