CLAIMS

1. A method for purifying full recombinant adeno-associated virus (rAAV) particles comprising:
    (a). providing an rAAV preparation comprising full rAAV particles and non-full rAAV particles;
    (b). loading the rAAV preparation in a loading buffer to a column comprising a chromatography medium, wherein the full rAAV particles have a higher binding affinity to the chromatography medium than the non-full particles, and the quantity of the full rAAV particles and non-full particles applied to the column exceeds the binding capacity of the chromatography medium, such that the non-full particles bound to the chromatography medium are displaced by the full rAAV particles into the flowthrough from the column; and
    (c). eluting the full rAAV particles bound to the chromatography medium with an elution buffer to obtain a purified preparation.
2. The method according to claim 1, wherein the chromatography medium is an ion exchange column chromatography medium, preferably an anion exchange chromatography medium.
3. The method according to claim 2, wherein the column chromatography medium is selected from the group consisting of Poros 50 HQ, Poros 50 D, Poros 50 PI, Capto ImpRes Q, CIMmultus™ QA Monolithic Column and Poros XQ, preferably Poros XQ.
4. A method for purifying full recombinant adeno-associated virus (rAAV) particles comprising:
    (a). providing an rAAV preparation comprising the full rAAV particles and non-full particles;
    (b). loading a first batch of the rAAV preparation in a loading buffer to a first column comprising a first chromatography medium, wherein the full rAAV particles have a higher binding affinity to the first chromatography medium than the non-full particles, and the quantity of the full rAAV particles and non-full particles applied to the first column exceeds the binding capacity of the first chromatography medium such that the non-full particles bound to the first

chromatography medium are displaced by the full rAAV particles into a first load flowthrough from the first column;

(c). loading the first load flowthrough to a second column comprising a second chromatography medium to obtain a loaded or partially loaded second column, wherein the second chromatography medium is the same type as the first chromatography medium;

(d). optionally, washing the first column with a wash buffer to obtain a washed first column;

(e). eluting the full rAAV particles bound to the first chromatography medium with an elution buffer to obtain a first eluate from the first column and an eluted first column, wherein the first eluate comprises an increased ratio of the full rAAV particles to the non-full rAAV particles;

(f). optionally, a second batch of the rAAV preparation in loading buffer is applied to the at least partially loaded second column, wherein the quantity of the full rAAV particles and non-full particles applied to the second column exceeds the binding capacity of the second chromatography medium such that the non-full particles bound to the second chromatography medium are displaced by the full rAAV particles into a second load flowthrough from the second column;

(g). optionally, loading the second load flowthrough from the second column to the eluted first column after step (e) to obtain a second loaded first column, and optionally washing second loaded first column before continuing to elute in step (e);

(h). optionally, washing the second column with a wash buffer to obtain a washed second column;

(i). eluting the full rAAV particles bound to the second chromatography medium with an elution buffer to obtain a second eluate and an eluted second column, wherein the second eluate comprises an increased ratio of the full rAAV particles to the non-full rAAV particles; and

(j). combining the first eluate and second eluate to produce a purified preparation of full rAAV particles.

5. The method according to claim 4, wherein the first chromatography medium and/or the second chromatography medium is an ion exchange column chromatography medium, preferably an anion exchange chromatography medium.
6. The method according to claim 4, wherein the steps (b) to (i) are performed with one or more cycles.
7. The method according to claim 4, wherein the second column is partially loaded after step (c).
8. The method according to claim 4, wherein the first column chromatography medium is selected from the group consisting of Poros 50 HQ, Poros 50 D, Poros 50 PI, Capto ImpRes Q, CIMmultus™ QA Monolithic Column and Poros XQ, preferably Poros XQ.

9.The method according to claim 1, wherein the loading buffer comprises at least one buffer selected from the group consisting of Tris, Bis-tris, Bis-tris propane, Tris acetate, ethanolamine, and phosphate.

10.The method according to claim 1, wherein the loading buffer comprises at least one salt of a cation selected from the group consisting of K(I), Li(I), Ca(II), Mg(II), Cu(II), Ba(II)), Co(II), Ni(II), Mn(II), Zn(II), Cd(II), Pb(II), Fe(III), Fe(II), Na(I), and NH_4^+.

11. The method according to claim 1, wherein the loading buffer has a pH of about 6-10.

12. The method according to claim 1, wherein the loading buffer comprises at least one surfactant. selected from the group consisting of poloxamer 188, polysorbate 80, polysorbate 20, NP-40, Triton X-100, and Triton CG-110.

13. The method according to claim 1, wherein the elution buffer comprises at least one salt of a cation selected from the group consisting of K(I), Li(I), Ca(II), Mg(II), Cu(II), Ba(II)), Co(II), Ni(II), Mn(II), Zn(II), Cd(II), Pb(II), Fe(III), Fe(II), Na(I), and NH_4^+.

14. The method according to claim 1, wherein the elution buffer comprises at least one buffer selected from the group consisting of Tris, Bis-tris, Bis-tris propane, Tris acetate, ethanolamine, and phosphate.

15. A method for purifying full recombinant adeno-associated virus (rAAV) particles comprising:

(a). providing an rAAV preparation comprising the full rAAV particles and non-full particles;

(b). loading the rAAV preparation in a loading buffer to a column comprising a chromatography medium, wherein the loading buffer comprises $CaCl_2$, and the full rAAV particles bind to the chromatography medium; and

(c). eluting the full rAAV particles bound to the chromatography medium with an elution buffer to obtain a purified preparation, wherein the elution buffer comprises $CaCl_2$.

16. The method according to claim 15, wherein the chromatography medium is an ion exchange column chromatography medium, preferably an anion exchange chromatography medium.

17. The method according to claim 16, wherein the column chromatography medium is selected from the group consisting of Poros 50 HQ, Poros 50 D, Poros 50 PI, Capto ImpRes Q, CIMmultus™ QA Monolithic Column, and Poros XQ, preferably Poros XQ.

18. The method according to claim 15, wherein the loading buffer comprises at least one salt of a cation selected from the group consisting of K(I), Li(I), Ca(II), Mg(II), Cu(II), Ba(II)), Co(II), Ni(II), Mn(II), Zn(II), Cd(II), Pb(II), Fe(III), Fe(II), Na(I), and NH_4^+.

19. The method according to claim 15, wherein the loading buffer comprises at least one buffer selected from the group consisting of Tris, Bis-tris, Bis-tris propane, Tris acetate, ethanolamine, and phosphate.

20. The method according to claim 15, wherein the elution buffer comprises at least one salt of a cation selected from the group consisting of K(I), Li(I), Ca(II), Mg(II), Cu(II), Ba(II)), Co(II), Ni(II), Mn(II), Zn(II), Cd(II), Pb(II), Fe(III), Fe(II), Na(I), and NH_4^+.