

CLAIMS – AUXILIARY REQUEST 88

1. A method to determine the serotype of an adeno-associated virus (AAV) particle comprising

- a) denaturing the AAV particle,
- b) directly subjecting the denatured AAV particle to liquid chromatography/mass spectrometry (LC/MS), and
- c) determining the masses of VP1, VP2 and VP3 of the AAV particle;

wherein the method is performed in the absence of a gel separation step; and

wherein the specific combination of masses of VP1, VP2 and VP3 are indicative of the AAV serotype,

optionally wherein the calculated masses of VP1, VP2 and VP3 are compared to the theoretical masses of VP1, VP2 and VP3 of one or more AAV serotypes.

~~2. A method of determining the heterogeneity of an AAV particle comprising~~

- ~~a) denaturing the AAV particle,~~
- ~~b) subjecting the denatured AAV particle to liquid chromatography/mass spectrometry/mass spectrometry (LC/MS/MS),~~
- ~~c) determining the masses of VP1, VP2 and VP3 of the AAV particle, and~~
- ~~d) comparing the masses of step c) with the theoretical masses of VP1, VP2 and VP3 of the AAV serotype;~~

~~wherein a deviation of one or more of the masses of VP1, VP2 or VP3 are indicative of the AAV capsid heterogeneity,~~

~~optionally wherein the heterogeneity comprises one or more of mixed serotypes, variant capsids, capsid amino acid substitutions, truncated capsids, or modified capsids.~~

~~32.~~ The method of ~~any one of claims 1–2,~~ wherein the AAV particle is denatured with acetic acid, guanidine hydrochloride and/or an organic solvent.

~~43.~~ The method of any one of claims ~~1–32,~~ wherein the liquid chromatography is reverse phase liquid chromatography, ~~size exclusion chromatography,~~ hydrophilic interaction liquid chromatography, or cation exchange chromatography.