



Supplementary Figure S2. Analysis of selected MWs for AAVrh8R/FVIII vector production and stability. **(A)** The stability of rAAV production by high-producing MWs. Vector productivity is shown for MW#287 (AAV8/FVIII-5.1), MW#35 (AAVrh8R/FVIII-5.1), and MW#163 (AAVrh8R/FVIII-5.4). Each was tested using two, three, and two independent experiments, respectively. MWs were passaged up to passage 20 or 26. In each section, the rAAV productivity (vg/mL) was quantified by qPCR using qPCR with FVIII-A2 primer/probe set. **(B)** Time-course for AAVrh8R/5.1 vector production using MW#35. Shaker cultures (250 mL) were infected with wild-type adenovirus, and samples were collected on days 2, 3, and 4. All values were tested in triplicates using qPCR with FVIII-A2 primer/probe set. Abbreviation: MW#35THAW, MW#35 that had undergone freezing and subsequent culture. MW#272, a medium producer, is shown for comparison. **(C)** Specific productivity for MW#35 at various production volumes. The specific production (SP) was measured by qPCR using FVIII-A2 primer/probe set and levels were normalized to cell number (n =number of independent measurements). The percentage of virus detected in the cell pellet (Cells) or culture media (CM) was measured (single experiment for each volume, values tested in triplicates). The specific productivity for a normal-size AAV2/SEAP-4.3 vector is shown for comparison.