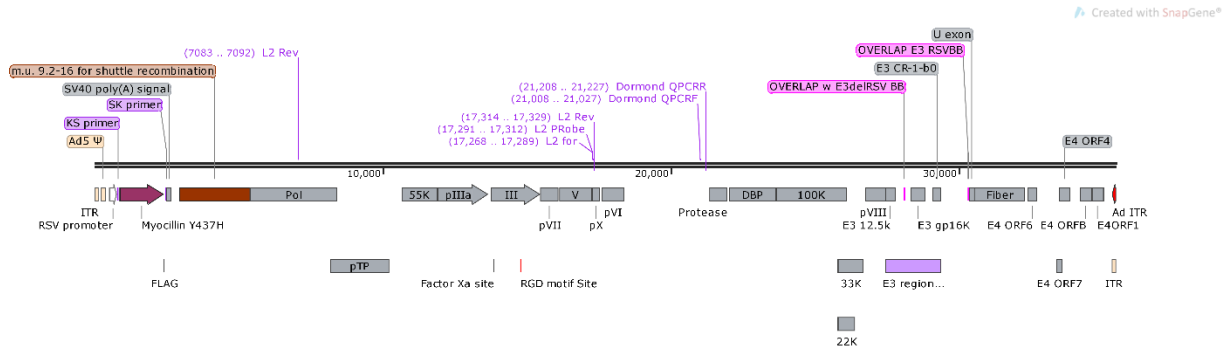


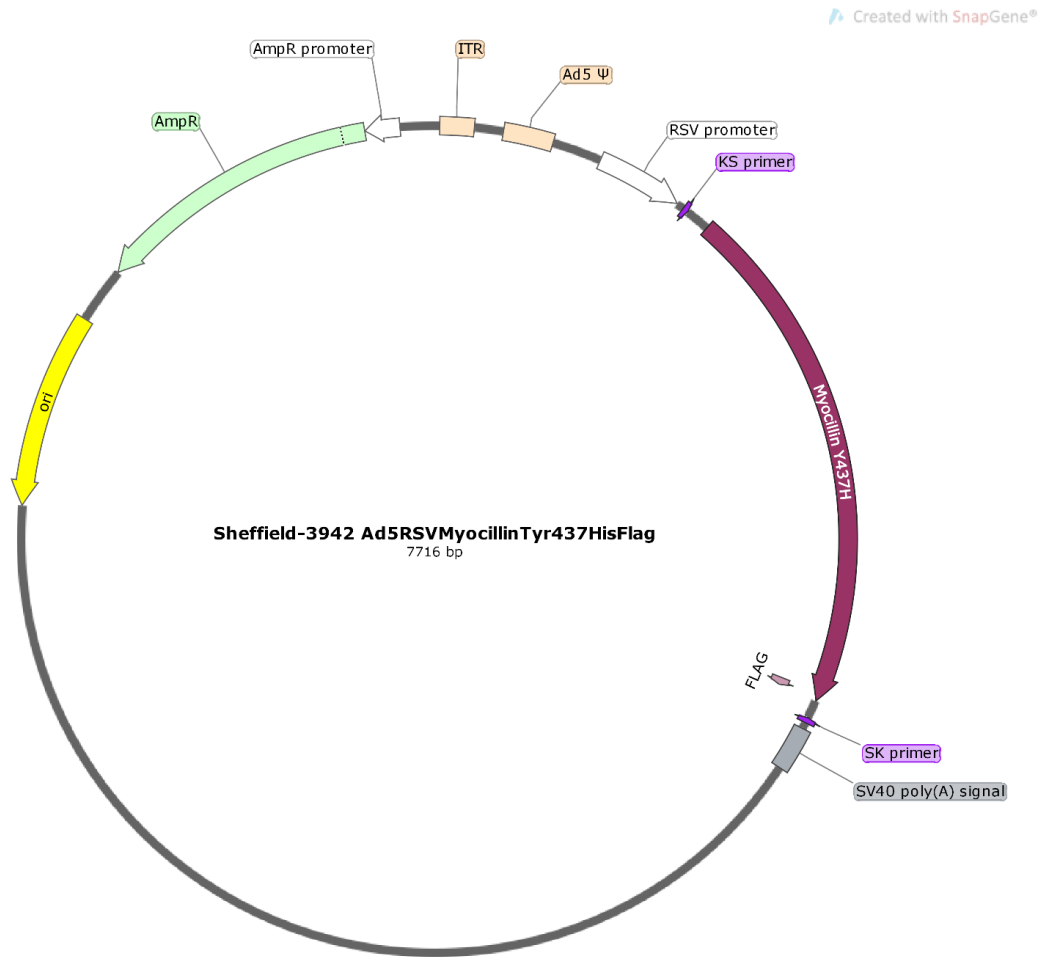
University of Iowa Biosafety Information for Sheffield-3942 Ad5RSVMycillinTyr437HisFlag

Viral DNA Map



Sheffield-3942 Ad5RSVMycillinTyr437HisFlag viral DNA
35,435 bp

Shuttle Plasmid Map and Sequence



Sheffield-3942 Ad5RSVMycillinTyr437HisFlag
7716 bp

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Vector Bio-safety Information

At the University of Iowa, all varieties of viral vectors produced at the Viral Vector Core are required to be handled at Biosafety Level 2 (BSL2). In animal studies, adenoviral vectors require ABL2 containment. Please check with your institution's Biosafety Officer to confirm local requirements

Adenovirus Background:

Adenoviruses are very important tool in basic research. They are used to identify proteins role in different biological processes both *in vivo* and *in vitro*. Virus construction is performed using the RapAd™ System developed by the University of Iowa GTVC (For description, refer to the article "*A simple method for the rapid generation of recombinant adenovirus vectors*" published in [Gene Therapy 7:1034-1038, 2000](#)). Adenovirus vectors prepared in the core are E1 and E3 deleted. They have a total E1a deletion (*m.u. 1.4 to 4.5) plus a partial E1b deletion (*m.u. 4.7 to 9.2). These deletions are what make the vector replication deficient. They also have a partial E3 deletion, 720bp for the sub360 backbone, a 1.6Kb deletion for the dl309 backbone and a 3.1Kb deletion for the total E3 deleted backbone.

Background on Virus production

The virus was made with our pacAd5(9.2-100)sub360 viral backbone. This backbone has a fully deleted E1a protein, a partially deleted E1b protein, and a partially deleted E3 protein to make the virus replication deficient. The vector preparations are purified by double CsCl protocol and dialyzed and stored in our A-195 buffer. All preparations are titered on HEK 293 cells using the Clontech Adeno-X titer kits and also tested for replication competent particles (RCA).

Recombination:

The recombinant adenoviruses can revert to wild type during virus production, thus packaging replication competent particles (RCA). For this reason, each new lot produced at the core is tested for the presence of RCA by immuno-staining.

References:

- **RapAd™ System:** Anderson RD, Haskell RE, Xia H, Roessler BJ, Davidson BL. "*A simple method*

for the rapid generation of recombinant adenovirus vectors". Gene Ther. 2000 Jun;7(12):1034-8

- **A195 Buffer:** [Evans RK](#), [Nawrocki DK](#), [Isopi LA](#), [Williams DM](#), [Casimiro DR](#), [Chin S](#), [Chen M](#), [Zhu DM](#), [Shiver JW](#), [Volkin DB](#). *Development of stable liquid formulations for adenovirus-based vaccines. J Pharm Sci.* 2004 Oct;93(10):2458-7

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