

## **Product datasheet**

# anti-AAV8 (intact particle) mouse monoclonal, ADK8, lyophilized, purified

#### Short overview

 Cat. No.
 610160

 Quantity
 50 μg

Concentration 50 µg/ml after reconstitution with 1 ml dist. water

#### **Product description**

Host Mouse
Antibody Type Monoclonal
Isotype IgG2a kappa
Clone ADK8

Immunogen AAV8 capsids

Formulation Lyophilized; after reconstitution with 1 ml dist. water solution contains 0.09% sodium azide and

0.5% BSA in PBS

**Binding affinity** KD value (AAV8) = <1.0E-12 M

KD value (AAVrh10) = <1.0E-12 M KD value (AAVrh74) = <1.0E-12 M Adeno-associated virus 8, AAV-8

Conjugate Unconjugated

**Purification** Affinity chromatography

**Storage before** 2-8°C until indicated expiry date

reconstitution

**Synomym** 

Storage after Up to 3 months at 2-8°C; long term storage in aliquots at -20°C; avoid freeze/thaw cycles

reconstitution

Intended use Research use only

**Application** Dot blot, ELISA, ICC/IF, IP, Neutralization assay **Reactivity** AAV3, AAV7, AAV8, AAVrh10, AAVrh74, Anc80

No reactivity AAV1, AAV11, AAV12, AAV2, AAV4, AAV5, AAV6, AAV9, AAVDJ

### **Applications**

**Dot Blot** 1:100-1:500 (0.1-0.5 μg/ml; non-denaturing conditions)

ELISA Assay dependent
Immunocytochemistry (ICC) Assay dependent
Immunoprecipitation (IP) Assay dependent

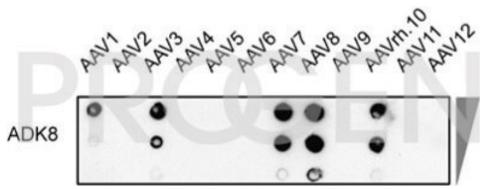
Neutralization Assay EC50 ~14 ng/ml (AAV3), ~4 ng/ml (AAV8) - assay dependent

### Background

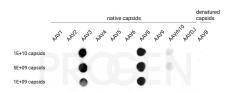
For characterization of different stages of infection and very useful for the analysis of the AAV assembly process. ADK8 specifically reacts with AAV1, AAV3, AAV7, AAV8, AAVrh10, AAVrh74 and Anc80, empty and full capsids. Recognizes a conformational epitope of assembled capsids. Predicted binding site: residues 586-LQQQNT-591 (Gurda et al. 2012). The antibody cannot be used for immunoblotting using denaturing conditions. The antibody is also useful for neutralizing experiments. Gurda, B. L. et al. Mapping a Neutralizing Epitope onto the Capsid of Adeno-Associated Virus Serotype 8. J. Virol. 86, 7739-7751 (2012).

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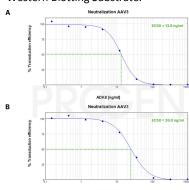
#### **Product images**



Dot blot with different AAV serotypes and mouse monoclonal anti-AAV8 antibody, clone ADK8 (Courtesy of Regina Heilbronn, Charité Universitätsmedizin Berlin, Mietzsch et al. Hum Gene Ther. 2014 Mar 1; 25(3):212-222)



Dot blot analysis of native AAV1-AAV9, AAVrh10, AAVDJ capsids (1E+09-1E+10 capsids) and denatured AAV8 capsids (1E+09-1E+10 capsids, denatured at 95°C for 10 min in sample buffer). The nitrocellulose membrane was blocked with 5% dry milk in PBST (PBS + 0.1% Tween 20) for 1 h at RT. The primary antibody anti-AAV8 (intact particle) mouse monoclonal, ADK8 (Cat. No. 610160) was diluted in blocking buffer (antibody concentration 100 ng/ml) and incubated for 1 h at RT. The secondary antibody goat anti-mouse IgG HRP was also diluted in blocking buffer (antibody concentration 200 ng/ml) and incubated for 1 h at RT. The bands were visualized by chemiluminescent detection using Pierce ECL Plus Western Blotting Substrate.



Neutralization of AAV3 with mouse monoclonal AAV8 antibody clone ADK8 (A) and human chimeric AAV8 antibody clone ADK8-h1 (B) by using AAV3-NanoLuc® viral particles from Promega. (A) anti-AAV8 (intact particle) mouse monoclonal, ADK8 (Cat. No. 610160) or (B) anti-AAV8, human chimeric, ADK8-h1 (Cat. No. 692318) were preincubated with AAV3-NanoLuc® viral particles for 30 min at RT at 300 rpm (antibody concentrations PROGEN Biotechnik GmbH | Maaßstraße 30 | D-69123 Heidelberg

0.2-3,000 ng/ml). HEK293 cells (100 µl) were plated at 200,000 cells/ml in DMEM + 1% FCS. Virus-antibody-mix (20 µl) was added to the cells and incubated for 16-24 h at 37°C. Extracellular NanoLuc Inhibitor and Nano-Glo® Live Cell Assay System (Promega) was added to the wells and incubated for 5 min at RT at 300 rpm. Luminescence was measured using an ID5-Reader and plotted with Softmax Pro 7.1 software to determine the EC50 values.

## References

Publication	Species	Application
François, A. et al. Accurate Titration of Infectious AAV Particles Requires Measurement of Biologically Active Vector	AAV8	ICC-IF
Genomes and Suitable Controls. Mol. Ther Methods Clin.		
Dev. 10, 223–236 (2018).		
Haar, J., Blazevic, D., Strobel, B., Kreuz, S. & Michelfelder, S. MSD-based assays facilitate a rapid and quantitative serostatus profiling for the presence of anti-AAV antibodies.  Mol. Ther Methods Clin. Dev. 25, 360–369 (2022).	AAV8	IA
Emmanuel, S. N., Mietzsch, M., Tseng, Y. S., Smith, J. K. & Agbandje-Mckenna, M. Parvovirus Capsid-Antibody Complex Structures Reveal Conservation of Antigenic Epitopes across the Family. Viral Immunol. 34, 3–17 (2021).	AAV8	binding region
Baatartsogt, N. et al. A sensitive and reproducible cell-based assay via secNanoLuc to detect neutralizing antibody against adeno-associated virus vector capsid. Mol. Ther Methods Clin. Dev. 22, 162–171 (2021).	AAV8	Neutralization
Gurda, B. L. et al. Mapping a Neutralizing Epitope onto the Capsid of Adeno-Associated Virus Serotype 8. J. Virol. 86, 7739-7751 (2012).	AAV8	ELISA,dot blot,ICC-IF,neutralization,epito pe mapping

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