

CaptureSelect™ HRP Anti-AAVX Conjugate

Catalog Number 7303522100

Pub. No. MAN0025684 Rev. A.0

Product description

CaptureSelect™ HRP Anti-AAVX Conjugate includes a 14-kDA camelid antibody fragment. The fragment specifically binds with high affinity to adeno-associated virus particles (AAV). It demonstrates good binding reactivity towards a variety of AAV serotypes, including AAV1 to AAV8, and AAVrh10. The antibody fragment is chemically conjugated to horseradish peroxidase (HRP) with a spacer of appropriate length to maintain full binding capacity of the antibody fragment.

Note: For assay development to measure AAV9 total capsids, we recommend CaptureSelect™ Biotin Anti-AAV9 Conjugate (Cat. No. [7103332100](#)) and CaptureSelect™ HRP Anti-AAV9 Conjugate (Cat. No. [7303332100](#)).

To quantify the total number of AAV capsids, the HRP anti-AAVX conjugate can be used with CaptureSelect™ Biotin Anti-AAVX Conjugate (Cat. No. [7103522100](#)) in Enzyme Linked Immuno-Sorbent Assays (ELISA). Sandwich ELISA set-ups can be designed using the biotin anti-AAVX conjugate as a capture reagent immobilized on a streptavidin-coated surface. The HRP anti-AAVX conjugate is then used for detection of bound AAV capsids (Figure 1).

The unique binding properties of the biotin and HRP anti-AAVX conjugates eliminates the need to incorporate different AAV serotype-specific antibodies for AAV assay development.

Contents and storage

Cat. No.	Amount	Storage
7303522100	100 µL ^[1] (0.5 mg/mL protein in PBS, pH 7.4 (50% (v/v) glycerol)	<1 month: 4°C >1 month: -30 to -5°C

^[1] The volume is sufficient for 50–100 ELISA plates.

Sandwich ELISA set up

Use the following recommended materials or their equivalents. See “Ordering Information” on page 2 for more information.

- (Wash) buffer—Pierce™ 20X PBS Tween™ -20 Buffer (diluted 1:20 in water, the solution yields 10 mM sodium phosphate, 0.15M NaCl, 0.05% Tween™ -20, pH 7.5)
- ELISA plate—Pierce™ Streptavidin Coated Plates, clear, 96-well
- HRP detection substrate—1-Step™ Ultra TMB-ELISA Substrate Solution
- Stop solution—1M sulfuric acid
- Standard plate reader with the appropriate filters

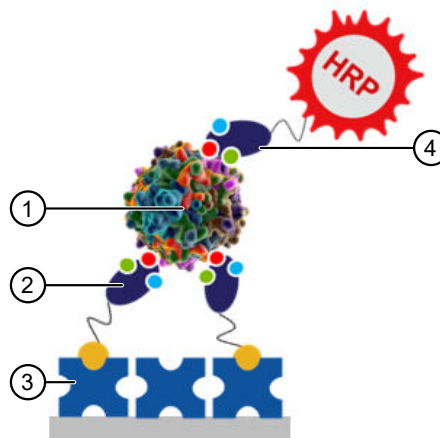


Figure 1 Anti-AAV sandwich ELISA

- ① AAV particle
- ② Anti-AAV biotin ligand (capture)
- ③ Streptavidin-coated ELISA plate
- ④ Anti-AAV HRP ligand (detection)

Determine the optimal working concentrations or dilutions for the steps below for each application.

Thermo Fisher Scientific does not supply standards for this application. Prepare a standard using purified AAV capsids, which most resemble the AAV vector of interest.

1. Prepare the CaptureSelect™ Biotin Anti-AAVX Conjugate (0.25–2 µg/mL in buffer).
2. Add 100 µL/well of the biotin conjugate to the ELISA plate. Incubate for 1 hour at room temperature on a microplate shaker.
3. Wash the ELISA plate 3 times with 200 µL/well of wash buffer.
4. Prepare a titration of the target AAV sample and reference standard in buffer (~10⁸ to 10¹¹ AAV capsids/mL).
5. Add 100 µL/well of the target AAV sample and reference standard to the ELISA plate. Incubate for 1 hour at room temperature on a microplate shaker.
6. Wash the ELISA plate 3 times with 200 µL/well of wash buffer.
7. Prepare CaptureSelect™ HRP Anti-AAVX Conjugate (1:5,000 to 1:20,000 dilution in buffer).
8. Add 100 µL/well of the HRP conjugate to the ELISA plate. Incubate for 1 hour at room temperature on a microplate shaker.
9. Wash the ELISA plate 3 times with 200 µL/well of wash buffer.
10. Add 100 µL/well of 1-Step™ Ultra TMB-ELISA Substrate Solution to the ELISA plate. Incubate for 5–10 minutes at room temperature on a microplate shaker.
11. Add 50 µL/well of 1M sulfuric acid to the ELISA plate to stop the reaction.
12. Measure the absorbance at 450 nm in a plate reader.

Example results

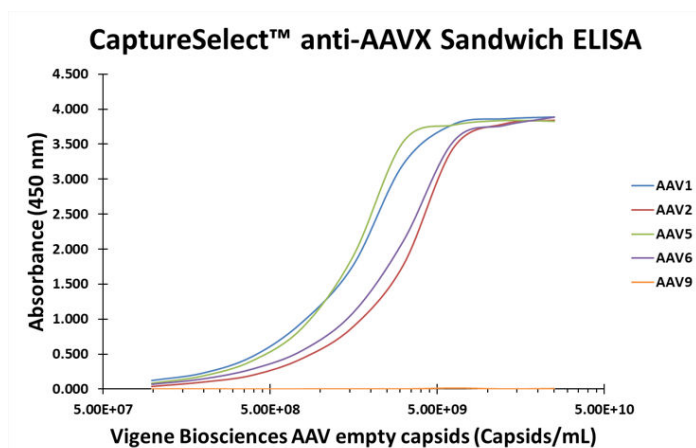


Figure 2 Anti-AAVX sandwich ELISA results with empty capsid reference standards (Vigene Biosciences)

Ordering Information

Product	Cat. No.
CaptureSelect™ HRP Anti-AAVX Conjugate	7303522100
CaptureSelect™ Biotin Anti-AAVX Conjugate	7103522100 (100 µg) 7103522500 (500 µg)
CaptureSelect™ HRP Anti-AAV9 Conjugate	7303332100
CaptureSelect™ Biotin Anti-AAV9 Conjugate	7103332100 (100 µg) 7103332500 (500 µg)
Pierce™ Streptavidin Coated Plates (pre-blocked with SuperBlock™ Blocking Buffer)	15124
1-Step™ Ultra TMB-ELISA Substrate Solution	34028
Pierce™ 20X PBS Tween™-20 Buffer	28352

For more information

For more information on CaptureSelect™ products and ligand leakage ELISA products, go to www.thermofisher.com/captureselect.

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 - Certificates of Analysis
 - Safety Data Sheets (SDSs; also known as MSDSs)

Note: For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

Limited product warranty

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References

Thermo Fisher Scientific, *Tech Tip #65, ELISA technical guide and protocols*



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Revision history: Pub. No. MAN0025684

Revision	Date	Description
A.0	01 September 2021	New document for CaptureSelect™ HRP Anti-AAVX Conjugate (Cat. No. 7303522100).

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