

Datasheet for ABIN6953714
Angiopoietin 1 ELISA Kit[Go to Product page](#)

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Overview

Quantity:	96 tests
Target:	Angiopoietin 1 (ANGPT1)
Reactivity:	Dog
Method Type:	Sandwich ELISA
Detection Range:	1.56 ng/mL - 100 ng/mL
Minimum Detection Limit:	1.56 ng/mL
Application:	ELISA

Product Details

Purpose:	The kit is a sandwich enzyme immunoassay for in vitro quantitative measurement of ANGPT1 in canine serum, plasma.
Sample Type:	Plasma, Serum
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	This assay has high sensitivity and excellent specificity for detection of Angiopoietin 1 (ANGPT1)
Sensitivity:	0.63 ng/mL
Components:	<ul style="list-style-type: none">• Pre-coated, ready to use 96-well strip plate, flat bottom• Plate sealer for 96 wells• Reference Standard• Standard Diluent

Product Details

- Detection Reagent A
- Assay Diluent A
- Reagent Diluent (if Detection Reagent is lyophilized)
- TMB Substrate
- Stop Solution
- Wash Buffer (30 x concentrate)
- Instruction manual

Target Details

Target:	Angiopoietin 1 (ANGPT1)
Abstract:	ANGPT1 Products
Pathways:	RTK Signaling, Glycosaminoglycan Metabolic Process

Application Details

Comment:	<p>Information on standard material:</p> <p>The standard might be recombinant protein or natural protein, that will depend on the specific kit. Moreover, the expression system is E.coli or yeast or mammal cell. There is 0.05% proclin 300 in the standard as preservative.</p> <p>Information on reagents:</p> <p>The stop solution used in the kit is sulfuric acid with concentration of 1 mol/L. And the wash solution is TBS. The standard diluent contains 0.02 % sodium azide, assay diluent A and assay diluent B contain 0.01% sodium azide. Some kits can contain is BSA in them.</p> <p>Information on antibodies:</p> <p>The provided antibodies and their host vary in different kits.</p>
Sample Volume:	100 µL
Assay Time:	3 h
Plate:	Pre-coated
Protocol:	<ol style="list-style-type: none">1. Prepare all reagents, samples and standards,2. Add 100µL standard or sample to each well. Incubate 1 hours at 37 °C,3. Aspirate and add 100µL prepared Detection Reagent A. Incubate 1 hour at 37 °C,4. Aspirate and wash 3 times,5. Add 90µL Substrate Solution. Incubate 10-20 minutes at 37 °C,

6. Add 50µL Stop Solution. Read at 450nm immediately.

Reagent Preparation:

1. Bring all kit components and samples to room temperature (18-25 °C) before use.
2. Standard - Reconstitute the Standard with 1.0 mL of Standard Diluent, keep for 10 minutes at room temperature, shake gently (not to foam). The concentration of the standard in the stock solution is 400 ng/mL. Firstly dilute the stock solution to 100 ng/mL and the diluted standard serves as the highest standard (100 ng/mL). Then prepare 7 tubes containing 0.5 mL Standard Diluent and use the diluted standard to produce a double dilution series. Mix each tube thoroughly before the next transfer. Set up 7 points of diluted standard such as 100 ng/mL, 50 ng/mL, 25 ng/mL, 12.5 ng/mL, 6.25 ng/mL, 3.12 ng/mL, 1.56 ng/mL, and the last microcentrifuge tube with Standard Diluent is the blank as 0 ng/mL.
3. Detection Reagent A - Briefly spin or centrifuge the stock Detection A before use. Dilute to the working concentration with working Assay Diluent A, respectively (1:100).
4. Wash Solution - Dilute 20 mL of Wash Solution concentrate (30x) with 580 mL of deionized or distilled water to prepare 600 mL of Wash Solution (1x).
5. TMB substrate - Aspirate the needed dosage of the solution with sterilized tips and do not dump the residual solution into the vial again.

Note:

1. Making serial dilution in the wells directly is not permitted.
 2. Prepare standard within 15 minutes before assay. Please do not dissolve the reagents at 37 °C directly.
 3. Please carefully reconstitute Standards or working Detection Reagent A according to the instruction, and avoid foaming and mix gently until the crystals are completely dissolved. To minimize imprecision caused by pipetting, use small volumes and ensure that pipettors are calibrated. It is recommended to suck more than 10µL for once pipetting.
 4. The reconstituted Standards, Detection Reagent A can be used only once.
 5. If crystals have formed in the Wash Solution concentrate (30x), warm to room temperature and mix gently until the crystals are completely dissolved.
 6. Contaminated water or container for reagent preparation will influence the detection result.
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Sample Preparation:

- It is recommended to use fresh samples without long storage, otherwise protein degradation and denaturation may occur in these samples, leading to false results. Samples should therefore be stored for a short period at 2 - 8 °C or aliquoted at -20 °C (≤1 month) or -80 °C (≤ 3 months). Repeated freeze-thaw cycles should be avoided. Prior to assay, the frozen samples should be slowly thawed and centrifuged to remove precipitates.
 - If the sample type is not specified in the instructions, a preliminary test is necessary to determine compatibility with the kit.
 - If a lysis buffer is used to prepare tissue homogenates or cell culture supernatant, there is a possibility of causing a deviation due to the introduced chemical substance. The recommended dilution factor is for reference only.
 - Please estimate the concentration of the samples before performing the test. If the values are not in the range of the standard curve, the optimal sample dilution for the particular experiment has to be determined. Samples should then be diluted with PBS (pH = 7.0-7.2).
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Application Details

Assay Procedure:	<ol style="list-style-type: none">1. Determine wells for diluted standard, blank and sample. Prepare 7 wells for standard, 1 well for blank. Add 100µL each of the dilutions of standard (see Reagent Preparation), blank and samples to the appropriate wells. Cover with a plate sealer. Incubate for 1 hour at 37°C.2. Remove the liquid from each well, do not wash.3. Add 100µL of Detection Reagent A Working Solution to each well, cover the wells with a plate sealer, and incubate at 37 °C for 1 hour.4. Aspirate the solution and add 350µL of 1x Wash Solution into each well using a squirt bottle, multichannel pipette, manifold dispenser, or automated washer and let it soak for 1-2 minutes. Remove the remaining liquid from all wells completely by tapping the plate on absorbent paper. Wash a total of 3 times. After the last wash, remove all remaining Wash Buffer by aspirating or decanting. Turn the plate over and blot it against absorbent paper.5. Add 90µL of Substrate Solution to each well. Cover with a new Plate Sealer. Incubate for 10-20 minutes at 37°C (do not exceed 30 minutes). Protect from light. The liquid will turn blue with the addition of Substrate Solution.6. Add 50µL of Stop Solution to each well. The liquid turns yellow due to the addition. Mix the liquid by tapping the side of the plate. If the color change does not appear even, gently tap the plate to mix thoroughly.7. Remove all water droplets and fingerprints from the bottom of the plate and make sure there are no bubbles on the surface of the liquid. Then run the microplate reader and immediately take a measurement at 450 nm.
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Assay Precision:	<p>Intra-assay Precision (Precision within an assay): 3 samples with low, middle and high level of target were tested 20 times on one plate, respectively.</p> <p>Inter-assay Precision (Precision between assays): 3 samples with low, middle and high level of target were tested on 3 different plates, 8 replicates in each plate.</p> <p>$CV(\%) = SD/mean \times 100$</p> <p>Intra-Assay: $CV < 10\%$</p> <p>Inter-Assay: $CV < 12\%$</p>
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Restrictions:	For Research Use only
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Handling

Precaution of Use:	The Stop Solution suggested for use with this kit is an acid solution. Wear eye, hand, face, and clothing protection when using this material.
Storage:	4 °C/-20 °C
Storage Comment:	<ol style="list-style-type: none">1. For unopened kit: All reagents should be stored according to the labels on the vials. The Standard, Detection Reagent A, Detection Reagent B, and 96-well Strip Plate should be stored at -20 °C upon receipt, while the other reagents should be stored at 4 °C.2. For opened kits: the remaining reagents must be stored according to the above storage

Handling

conditions. In addition, please return the unused wells to the foil pouch containing the desiccant and seal the foil pouch with the zipper.

Expiry Date: 6 months

Images

ELISA

Image 1. Typical standard curve

