# abcam

## Product datasheet

## Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] - BSA and Azide free ab216646

Recombinant RabMAb

**10 References** 12 Images

Overview

**Product name** Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] - BSA and Azide free

**Description** Rabbit monoclonal [CAN-R9(IHC)-56-2] to Wilms Tumor Protein - BSA and Azide free

**Host species** Rabbit

Specificity Expression levels of the target protein vary with sample type and some optimisation may be

**Tested applications** Suitable for: IHC-P, WB, ICC/IF, Flow Cyt (Intra)

Unsuitable for: IP

Species reactivity Reacts with: Mouse, Human

**Immunogen** Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: K562 and Ramos cell lysate. IHC-P: Human kidney and Wilms tumor tissues. ICC/IF: K562

cells. Flow Cyt (intra): K562 cells.

**General notes** ab216646 is the carrier-free version of ab89901.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for

increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free production

For more information see here.

Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

## **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.20

Constituent: PBS

Carrier free Yes

Purity Protein A purified

**Clonality** Monoclonal

Clone number CAN-R9(IHC)-56-2

**Isotype** IgG

## **Applications**

#### The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab216646 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.  See IHC antigen retrieval protocols.
WB		Use at an assay dependent concentration. Predicted molecular weight: 55 kDa.
ICC/IF		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration. <u>ab199376</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

**Application notes** 

Is unsuitable for IP.

## **Target**

#### **Function**

Transcription factor that plays an important role in cellular development and cell survival. Regulates the expression of numerous target genes, including EPO. Plays an essential role for development of the urogenital system. Recognizes and binds to the DNA sequence 5'-CGCCCCGC-3'. It has a tumor suppressor as well as an oncogenic role in tumor formation. Function may be isoform-specific: isoforms lacking the KTS motif may act as transcription factors. Isoforms containing the KTS motif may bind mRNA and play a role in mRNA metabolism or

splicing. Isoform 1 has lower affinity for DNA, and can bind RNA.

#### Tissue specificity

#### Involvement in disease

Expressed in the kidney and a subset of hematopoietic cells.

Defects in WT1 are the cause of Frasier syndrome (FS) [MIM:136680]. FS is characterized by a slowly progressing nephropathy leading to renal failure in adolescence or early adulthood, male pseudohermaphroditism, and no Wilms tumor. As for histological findings of the kidneys, focal glomerular sclerosis is often observed. There is phenotypic overlap with Denys-Drash syndrome. Inheritance is autosomal dominant.

Defects in WT1 are the cause of Wilms tumor 1 (WT1) [MIM:194070]. WT is an embryonal malignancy of the kidney that affects approximately 1 in 10'000 infants and young children. It occurs both in sporadic and hereditary forms.

Defects in WT1 are the cause of Denys-Drash syndrome (DDS) [MIM:194080]. DDS is a typical nephropathy characterized by diffuse mesangial sclerosis, genital abnormalities, and/or Wilms tumor. There is phenotypic overlap with WAGR syndrome and Frasier syndrome. Inheritance is autosomal dominant, but most cases are sporadic.

Defects in WT1 are the cause of nephrotic syndrome type 4 (NPHS4) [MIM:256370]. A renal disease characterized clinically by proteinuria, hypoalbuminemia, hyperlipidemia and edema. Kidney biopsies show non-specific histologic changes such as focal segmental glomerulosclerosis and diffuse mesangial proliferation. Some affected individuals have an inherited steroid-resistant form and progress to end-stage renal failure. Most patients with NPHS4 show diffuse mesangial sclerosis on renal biopsy, which is a pathologic entity characterized by mesangial matrix expansion with no mesangial hypercellularity, hypertrophy of the podocytes, vacuolized podocytes, thickened basement membranes, and diminished patency of the capillary lumen.

Defects in WT1 are a cause of Meacham syndrome (MEACHS) [MIM:608978]. Meacham syndrome is a rare sporadically occurring multiple malformation syndrome characterized by male pseudohermaphroditism with abnormal internal female genitalia comprising a uterus and double or septate vagina, complex congenital heart defect and diaphragmatic abnormalities.

Note=A chromosomal aberration involving WT1 may be a cause of desmoplastic small round cell tumor (DSRCT). Translocation t(11;22)(p13;q12) with EWSR1.

## Sequence similarities

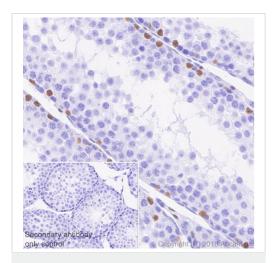
Belongs to the EGR C2H2-type zinc-finger protein family.

Contains 4 C2H2-type zinc fingers.

## **Cellular localization**

Nucleus. Cytoplasm. Shuttles between nucleus and cytoplasm; Nucleus > nucleoplasm and Nucleus speckle.

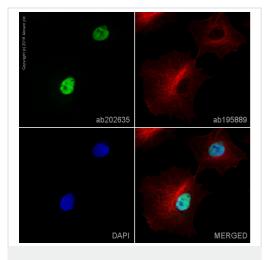
#### **Images**



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] - BSA and Azide free (ab216646)

Ab89901 staining Wilms Tumor Protein in paraffin-embedded Mouse testis tissue sections by Immunohistochemistry. Antigen retrieval was by heat mediation using citrate buffer (pH 6.0). Samples were incubated with primary antibody at 1:500 dilution (0.49  $\mu$ g/ml). A ready to use Goat Anti-Rabbit IgG H&L (HRP) was used as the secondary antibody. Nuclear staining on Sertoli cells in mouse testis (PMID: 21863216).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab89901).



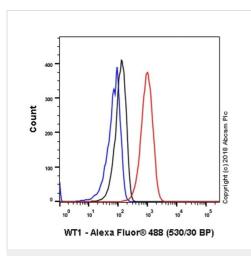
Immunocytochemistry/ Immunofluorescence - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] -BSA and Azide free (ab216646)

Clone CAN-R9(IHC)-56-2 (ab216646) has been successfully conjugated by Abcam. This image was generated using Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] (Alexa Fluor<sup>®</sup> 488). Please refer to **ab202635** for protocol details.

ab202635 staining Wilms Tumor Protein in HepG2 cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab202635 at 1/200 dilution (shown in green) and ab195889, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 594), at 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

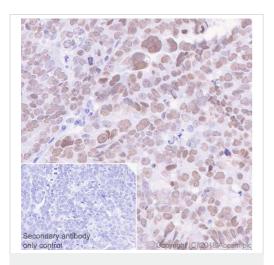
This product also gave a positive signal under the same testing conditions in HepG2 cells fixed with 100% methanol (5min).



Flow Cytometry (Intracellular) - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] - BSA and Azide free (ab216646)

Intracellular Flow Cytometry analysis of K-562 (Human chronic myelogenous leukemia lymphoblast) cells labeling Wilms Tumor Protein with <a href="mailto:ab89901">ab89901</a> at 1/200 dilution (0.1µg)/ Red. Cells were fixed with 4% paraformaldehyde and permeabilized with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor<sup>®</sup> 488, <a href="mailto:ab150077">ab150077</a>) at 1/2000 dilution was used as the secondary antibody. Rabbit monoclonal IgG (<a href="mailto:ab172730">ab172730</a>) / Black was used as the isotype control. Cells without incubation with primary antibody and secondary antibody / Blue was used as the unlabeled control.

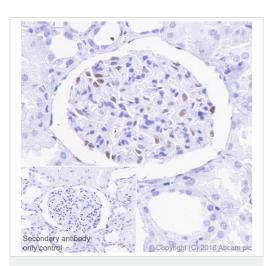
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab89901).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] - BSA and Azide free (ab216646)

Ab89901 staining Wilms Tumor Protein in paraffin-embedded Human ovarian serous adenocarcinoma tissue sections by Immunohistochemistry. Antigen retrieval was by heat mediation using citrate buffer (pH 6.0). Samples were incubated with primary antibody at 1:500 dilution (0.49 µg/ml). A ready to use Goat Anti-Rabbit IgG H&L (HRP) was used as the secondary antibody. Nuclear staining on human ovarian serous adenocarcinoma (PMID: 11939727).

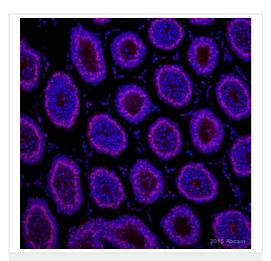
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab89901).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] - BSA and Azide free (ab216646)

Ab89901 staining Wilms Tumor Protein in paraffin-embedded Human kidney tissue sections by Immunohistochemistry. Antigen retrieval was by heat mediation using citrate buffer (pH 6.0). Samples were incubated with primary antibody at 1:500 dilution (0.49  $\mu$ g/ml). A ready to use Goat Anti-Rabbit lgG H&L (HRP) was used as the secondary antibody. Nuclear staining on human kidney glomerulus (PMID: 12898605).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab89901).

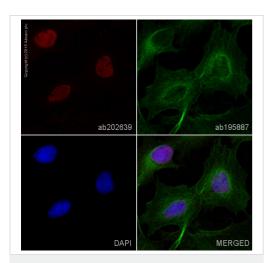


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] - BSA and Azide free (ab216646)

This image is courtesy of an anonymous Abreview.

Immunohistochemical analysis of fromaldehyde fixed, paraffin embedded rat testis tissue sections, labelling Wilms Tumor Protein using <u>ab89901</u>. Heat mediated antigen retrival was performed using 10 mM Sodium Citrate and 0.05% Tween 20. Tissue sections were incubated with <u>ab89901</u> at a 1/50 dilution for 12 hours at 4°C. The tissues were blocked with 10% Serum for 30 minutes at 25°C. The secondary used was a Donkey CY3<sup>®</sup> conjugate at a 1/200 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab89901).

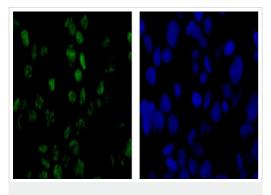


Immunocytochemistry/ Immunofluorescence - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] -BSA and Azide free (ab216646)

Clone CAN-R9(IHC)-56-2 (ab216646) has been successfully conjugated by Abcam. This image was generated using Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] (Alexa Fluor<sup>®</sup> 647). Please refer to <u>ab202639</u> for protocol details.

**ab202639** staining Wilms Tumor Protein in HepG2 cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with **ab202639** at 1/200 dilution (shown in red) and **ab195887**, Mouse monoclonal to alpha Tubulin (Alexa Fluor<sup>®</sup> 488), at 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

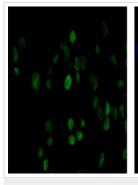
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunocytochemistry/ Immunofluorescence - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] -BSA and Azide free (ab216646)

Immunocytochemsitry/Immunofluorescence analysis of K562 cells labelling Wilms Tumor Protein (green) with purified <u>ab89901</u> at 1/50. Cells were fixed with 4% paraformaldehyde. An Alexa Fluor<sup>®</sup> 488-conjugated goat anti-rabbit lgG (1/200) was used as the secondary antibody. Counterstained with DAPI (blue).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab89901).

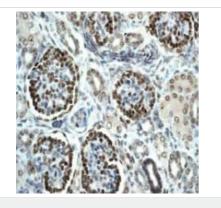




Immunocytochemistry/ Immunofluorescence - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] -BSA and Azide free (ab216646)

Immunocytochemsitry/Immunofluorescence analysis of K562 cells labelling Wilms Tumor Protein (green) with unpurified <u>ab89901</u> at 1/50. Cells were fixed with 4% paraformaldehyde. An Alexa Fluor<sup>®</sup> 488-conjugated goat anti-rabbit lgG (1/200) was used as the secondary antibody. Counterstained with DAPI (blue).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab89901).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] - BSA and Azide free (ab216646)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human fetal tissue labelling Wilms Tumor Protein with unpurified **ab89901** at 1/250.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab89901).

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-56-2] - BSA and Azide free (ab216646)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human Wilms tumor tissue labelling Wilms Tumor Protein with unpurified <u>ab89901</u> at 1/250.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab89901).

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.







Confirmed specificity

Ethical standards Animal-free production

Anti-Wilms Tumor Protein antibody [CAN-R9(IHC)-

56-2] - BSA and Azide free (ab216646)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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