

## Technical Data Sheet

## BV421 Rat Anti-Pax-5

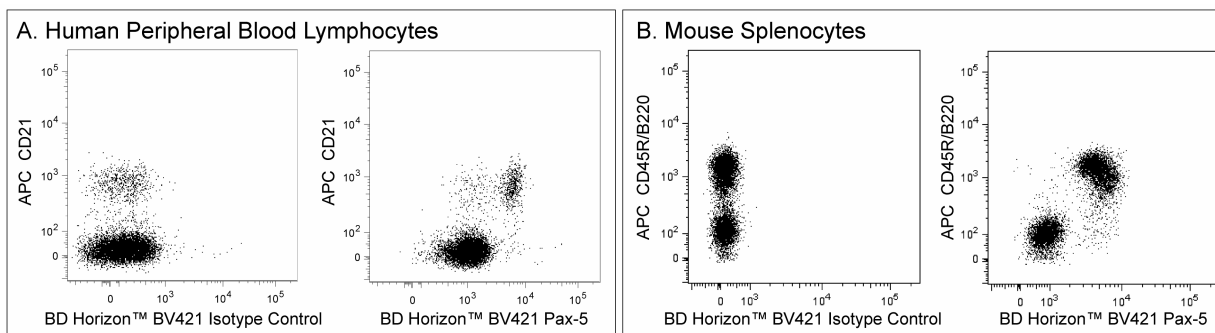
## Product Information

<b>Material Number:</b>	<b>563190</b>
<b>Alternate Name:</b>	Pax5, PAX5, KLP, BSAP, EBB-1; B-cell-specific transcription factor
<b>Size:</b>	50 tests
<b>Vol. per Test:</b>	5 µl
<b>Clone:</b>	1H9
<b>Immunogen:</b>	Recombinant Mouse Pax-5 protein containing aa 154-284
<b>Isotype:</b>	Rat IgG2a, κ
<b>Reactivity:</b>	QC Testing: Mouse Tested in Development: Human
<b>Storage Buffer:</b>	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

## Description

The 1H9 monoclonal antibody clone specifically binds to human and mouse Paired box protein Pax-5. Pax-5 is a ~50 kDa protein that is also known as B-cell-specific transcription factor and B cell specific activator protein (BSAP). Pax-5 is a member of the paired box (Pax) family of transcription factors. It is expressed in pro-B, pre-B and mature B cells. Through the Groucho family of co-repressors, Pax-5 likely functions as a transcriptional repressor of non-B-lymphoid genes during the B cell commitment process. In the early stages of B cell development, Pax-5 influences the expression of several B-cell-specific genes, such as CD19 and CD20 and maintains B cell identity. Pax-5 suppression is involved in the upregulation of Blimp-1 leading to the development of Pax-5-negative plasma cells. Pax-5 mRNA is transiently detected in the mesencephalon and spinal cord during embryogenesis. Expression then shifts to the fetal liver and correlates with the onset of B lymphopoiesis. Altered forms and expression patterns of Pax-5 have been associated with some lymphoid and nonlymphoid cancers.

The antibody was conjugated to BD Horizon™ BV421 which is part of the BD Horizon™ Brilliant Violet™ family of dyes. With an Ex Max of 407-nm and Em Max at 421-nm, BD Horizon™ BV421 can be excited by the violet laser and detected in the standard Pacific Blue™ filter set (eg, 450/50-nm filter). BD Horizon™ BV421 conjugates are very bright, often exhibiting a 10 fold improvement in brightness compared to Pacific Blue™ conjugates.



**Multicolor flow cytometric analysis of Pax-5 expression in human and mouse leucocytes.** Human peripheral blood mononuclear cells (PBMC) and BALB/c mouse splenic leucocytes were fixed and permeabilized using the BD Pharmingen™ Transcription Factor Buffer Set (Cat. No. 562574/562725). The cells were then stained with either BD Horizon™ BV421 Rat Anti-Mouse Pax-5 antibody (Cat. No. 563190) or BD Horizon™ BV421 Rat IgG2a, κ Isotype Control (Cat. No. 562602). The PBMC were counterstained with APC Mouse Anti-Human CD21 antibody (Cat. No. 561767/561357/559867). The mouse splenic leucocytes were counterstained with APC Rat Anti-Mouse CD45R/B220 antibody (Cat. No. 553092/561880). Flow cytometric analysis was performed using a BD LSR™ II Flow Cytometer System.

**Panel A. Human Peripheral Blood Lymphocytes:** The two-color flow cytometric dot plots show the correlated expression patterns of Ig isotype control staining (Left Plot) and Pax-5 (Right Plot) versus CD21 for events with the forward and side light-scatter characteristics of intact human peripheral blood lymphocytes.

**Panel B. Mouse Splenocytes:** The dot plots show the coexpression patterns of Ig isotype control staining (Left Plot) or Pax-5 (Right Plot) versus CD45R/B220 for events with the light-scatter characteristics of intact mouse splenocytes.

## Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated with BD Horizon™ BV421 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV421 were removed.

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## Application Notes

### Application

Intracellular staining (flow cytometry)

Routinely Tested

### Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 ml	(none)
562602	BV421 Rat IgG2a, $\kappa$ Isotype Control	50 $\mu$ g	R35-95
562574	Transcription Factor Buffer Set	100 tests	(none)
562725	Transcription Factor Buffer Set	25 tests	(none)
555899	Lysing Buffer	100 ml	(none)

### Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use  $1 \times 10^6$  cells in a 100- $\mu$ l experimental sample (a test).
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. An isotype control should be used at the same concentration as the antibody of interest.
4. Please refer to [www.bdbiosciences.com/pharming/en/protocols](http://www.bdbiosciences.com/pharming/en/protocols) for technical protocols.
5. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
7. Pacific Blue™ is a trademark of Molecular Probes, Inc., Eugene, OR.
8. Brilliant Violet™ 421 is a trademark of Sirigen.

### References

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Kallies A, Hasbold J, Fairfax K, et al. Initiation of plasma-cell differentiation is independent of the transcription factor Blimp-1. *Immunity.* 2007; 26(5):555-566. (Immunogen: Western blot)

Klein U, Tu Y, Stolovitzky GA, et al. Transcriptional analysis of the B cell germinal center reaction. *Proc Natl Acad Sci U S A.* 2003; 100(5):2639-2644. (Biology)

McManus S, Ebert A, Salvaggio G, Medvedovic J, Sun Q, Tamir I, Jaritz M, Tagoh H, Busslinger M. The transcription factor Pax5 regulates its target genes by recruiting chromatin-modifying proteins in committed B cells. *EMBO J.* 2011; 30(12):2388-2404. (Clone-specific: Western blot)

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