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Product datasheet

Human IL-1 Beta ELISPOT Kit ab62924

Overview

Product name	Human IL-1 Beta ELISPOT Kit
Detection method	Colorimetric
Sample type	Suspension cells
Assay type	Sandwich (qualitative)
Assay duration	Multiple steps standard assay
Species reactivity	**Reacts with:** Human
Product overview	The ELISPOT assay is designed to enumerate cytokine producing cells in a single cell suspension. This method has the advantage of requiring a minimum of in-vitro manipulations allowing cytokine production analysis as close as possible to in-vivo conditions in a highly specific way. This technique is designed to determine the frequency of cytokine producing cells under a given stimulation, and the follow-up of such frequency during a treatment and/or a pathological state. Elispot assay constitutes an ideal tool in the TH1 / TH2 response, vaccine development, viral infection monitoring and treatment, cancerology, infectious diseases, autoimmune diseases and transplantation. Abcam Elispot assay is based on sandwich immuno-enzyme technology. Cell secreted cytokines or soluble molecules are captured by coated antibodies avoiding diffusion in supernatant, protease degradation or binding on soluble membrane receptors. After cell removal, the captured cytokines are revealed by tracer antibodies and appropriate conjugates. **Principal:** After cell stimulation, locally produced cytokines are captured by a specific monoclonal antibody. After cell lysis, trapped cytokine molecules are revealed by a secondary biotinylated detection antibody, which is in turn recognised by streptavidin conjugated to alkaline phosphatase. PVDF-bottomed-well plates are then incubated with BCIP/NBT substrate. Colored "purple" spots indicate cytokine production by individual cells. Recognizes natural (pro and mature) human IL1 Beta
Tested applications	**Suitable for:** ELISpot
Platform	Microplate

Properties

Storage instructions	Store at +4°C. Please refer to protocols.

Components	1 x 96 tests	5 x 96 tests
Bovine Serum Albumin	1 x 200mg	1 x 1g
IL-1β pre-coated 96-well PDVF-bottomed plates	1 unit	5 units
IL-1ß Biotinylated Detection antibody	1 vial	1 vial
Ready-to-use BCIP/NBT substrate buffer	1 x 10ml	1 x 50ml
Streptavidin - Alkaline Phosphatase conjugated	1 x 10µl	1 x 50µl

Function	Potent proinflammatory cytokine. Initially discovered as the major endogenous pyrogen, induces prostaglandin synthesis, neutrophil influx and activation, T-cell activation and cytokine production, B-cell activation and antibody production, and fibroblast proliferation and collagen production. Promotes Th17 differentiation of T-cells.
Tissue specificity	Expressed in activated monocytes/macrophages (at protein level).
Sequence similarities	Belongs to the IL-1 family.
Post-translational modifications	Activation of the IL1B precursor involves a CASP1-catalyzed proteolytic cleavage. Processing and secretion are temporarily associated.
Cellular localization	Cytoplasm, cytosol. Lysosome. Secreted, exosome. Cytoplasmic vesicle, autophagosome. Secreted. The precursor is cytosolic. In response to inflammasome-activating signals, such as ATP for NLRP3 inflammasome or bacterial flagellin for NLRC4 inflammasome, cleaved and secreted. IL1B lacks any known signal sequence and the pathway(s) of its secretion is(are) not yet fully understood (PubMed:24201029). On the basis of experimental results, several unconventional secretion mechanisms have been proposed. 1. Secretion via secretory lysosomes: a fraction of CASP1 and IL1B precursor may be incorporated, by a yet undefined mechanism, into secretory lysosomes that undergo Ca(2+)-dependent exocytosis with release of mature IL1B (PubMed:15192144). 2. Secretory autophagy: IL1B-containing autophagosomes may fuse with endosomes or multivesicular bodies (MVBs) and then merge with the plasma membrane releasing soluble IL1B or IL1B-containing exosomes (PubMed:24201029). However, autophagy impacts IL1B production at several levels and its role in secretion is still controversial. 3. Secretion via exosomes: ATP-activation of P2RX7 leads to the formation of MVBs containing exosomes with entrapped IL1B, CASP1 and other inflammasome components. These MVBs undergo exocytosis with the release of exosomes. The release of soluble IL1B occurs after the lysis of exosome membranes (By similarity). 4. Secretion by microvesicle shedding: activation of the ATP receptor P2RX7 may induce an immediate shedding of membrane-derived microvesicles containing IL1B and possibly inflammasome components. The cytokine is then released in the extracellular compartment after microvesicle lysis (PubMed:11728343). 5. Release by translocation through permeabilized plasma membrane. This may occur in cells undergoing pyroptosis due to sustained activation of the inflammasome (By similarity). These mechanisms may not be not mutually exclusive.

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab62924 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
ELISpot		Use at an assay dependent dilution.

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