Datasheet for ABIN625575

Human Cytokine Antibody Array G1000

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Overview

Quantity:	4 samples
Reactivity:	Human
Method Type:	Sandwich ELISA
Application:	Antibody Array (AA)

Product Details

Purpose:	RayBio® Human Cytokine Antibody Array G Series 1000 kit with Accessories, a combination of RayBio® Human Cytokine Antibody Arrays G Series 6 & 7 (1 glass chip each per kit, 2 chips per kit, 8 sub-arrays per chip), for simultaneous detection of 120 Cytokin
Brand:	RayBio®
Sample Type:	Serum, Plasma, Cell Culture Supernatant, Cell Lysate, Tissue Lysate
Analytical Method:	Semi-Quantitative
Detection Method:	Fluorometric
Specificity:	Detects:Adiponectin(ACRP30), AgRP, Amphiregulin, Angiogenin, Angiopoietin-2, Axl, BDNF, beta-NGF, Betacellulin (BTC), bFGF, BLC (CXCL13), BMP-4, BMP-6, CCL28 (MEC), CK beta 8-1 (CCL23), CNTF, CTACK(CCL27), Dtk, EGF, EGFR, ENA-78 (CXCL5), Eotaxin-1 (CCL11), Eotaxin-2 (MPIF-2/CCL24), Eotaxin-3 (CCL26), Fas (TNFRSF6/Apo-1), FGF-4, FGF-6, FGF-7(KGF), FGF-9, Flt-3 Ligand, Fractalkine (CX3CL1), GCP-2 (CXCL6), GCSF, GDNF, GITR (TNFRSF18), GITR Ligand (TNFSF18), GM-CSF, gp130, GRO alpha (CXCL1), GRO alpha/beta/gamma, HCC-4 (CCL16), HGF, I-309 (TCA-3/CCL1), I-TAC(CXCL11), ICAM-1 (CD54), ICAM-3 (CD50), IFN-gamma, IGF-1, IGF-1 R, IGFBP-1, IGFBP-2, IGFBP-3, IGFBP-4, IGFBP-6, IL-1 alpha (IL-1 F1), IL-1 beta (IL-1 F2), IL-1 R1, IL-1 R4 (ST2), IL-1 ra (IL-1 F3), IL-10, IL-11, IL-12 p40, IL-12 p70, IL-13, IL-

Product Details

	15, IL-16, IL-17A, IL-2, IL-2 R alpha, IL-3, IL-4, IL-5, IL-6, IL-6 R, IL-7, IL-8 (CXCL8), Leptin, Light (TNFSF14), Lymphotactin (XCL1), M-CSF, MCP-1 (CCL2), MCP-2 (CCL8), MCP-3 (MARC/CCL7), MCP-4 (CCL13), MDC(CCL22), MIF, MIG (CXCL9), MIP-1 alpha (CCL3), MIP-1 beta (CCL4), MIP-1 delta (CCL15), MIP-3 alpha (CCL20), MIP-3 beta (CCL19), MSP alpha/beta, NAP-2 (PPBP/CXCL7), NT-3, NT-4, Oncostatin M, Osteoprotegerin (TNFRSF11B), PARC (CCL18), PDGF-BB, PLGF, RANTES (CCL5), SCF, SDF-1 alpha (CXCL12 alpha), TARC (CCL17), TECK (CCL25), TGF beta 1, TGF beta 3, Thrombopoietin (TPO), TIMP-1, TIMP-2, TNF alpha, TNF beta (TNFSF1B), TNF RI (TNFRSF1A), TNF RII (TNFRSF1B), TRAIL R3 (TNFRSF10C), TRAIL R4 (TNFRSF10D), uPAR, VEGF-A, VEGF-D
Components:	- RayBio® Antibody Array glass slide (4 or 8 arrays per slide) - Biotinylated Detection Antibodies - Streptavidin-conjugated HiLytePlus™ 555 Fluor - Blocking Buffer - 20X Wash Buffer I - 20X Wash Buffer II - 2X Cell Lysis Buffer - G-Series Antibody Array accessories* - Manual - Accessories include: 16-well incubation chamber with gasket, protective cover, snap-on sides, adhesive film
Material not included:	Distilled or deionized water Small plastic boxes or containers Pipettors, pipette tips and other common lab consumables Orbital shaker or oscillating rocker Aluminum foil Gene microarray scanner or similar laser fluorescence scanner

Target Details

Background:	Cytokines play an important role in innate immunity, apoptosis, angiogenesis, cell growth and differentiation. They are involved in interactions between different cell types, cellular responses to environmental conditions, and maintenance of homeostasis. In addition, cytokines are also involved in most disease processes, including cancer and cardiac diseases.

Application Details

Application Notes:	Completely cover array area with sample or buffer during incubation. Avoid foaming during incubation steps. Perform all incubation and wash steps under gentle rocking or rotation. Cover the incubation chamber with adhesive film during incubation, particularly when incubation is more than 2 hours or <70 µl of sample or reagent is used. Several incubation steps such as step 6 (blocking), step 7 (sample incubation), step 10 (detection antibody incubation), or step 13 (Cy3 equivalent dyestreptavidin incubation) may be done overnight at 4°C. Please make sure to cover the incubation chamber tightly to prevent evaporation.
Comment:	The G-Series arrays feature fluorescent signal detection. The antibodies are spotted on glass slide solid supports and require a laser scanner for data collection. All G-Series arrays work on the sandwich ELISA principle, utilizing a matched pair of antibodies: an immobilized capture antibody and a corresponding biotinylated detection antibody.
Sample Volume:	50-100 µL
Assay Time:	6 h
Plate:	Glass Slide
Protocol:	- Dry the glass slide - Prepare Standards - Block array surface - Incubate with samples - Incubate with Biotinylated Detection Antibody Cocktail - Incubate with Streptavidin-Conjugated Fluor - Disassemble the glass slide - Scan with a gene microarray laser scanner - Perform densitometry and analysis
Sample Preparation:	Use serum-free conditioned media if possible. If serum-containing conditioned media is required, it is highly recommended that complete medium be used as a control since many types of sera contains cytokines. We recommend the following parameters for your samples: 50 to 100 µl of original or diluted serum, plasma, cell culture media, or other body fluid, or 50-500 µg/ml of protein for cell and tissue lysates. If you experience high background or if the fluorescent signal intensities exceed the detection range, further dilution of your sample is recommended.
Assay Procedure:	Take out the glass slide from the box, and let it equilibrate to room temperature inside the sealed plastic bag for 20-30 minutes. Remove slide from the plastic bag, peel off the cover film, and let it air dry for another 1-2 hours.

Blocking & Incubation

1. Add 100 µl Sample Diluent into each well and incubate at room temperature for 30 minutes to block slides.

2. Decant buffer from each well. Add 100 µl of sample to each well. Incubate arrays at room temperature for 1-2 hour.

3. Decant the samples from each well, and wash 5 times (5 min each) with 150 µl of 1X Wash Buffer I at room temperature with gentle shaking. Completely remove wash buffer in each wash step. Dilute 20x Wash Buffer I with H2O.

4. Decant the 1x Wash Buffer I from each well, wash 2 times (5 min each) with 150 µl of 1X Wash Buffer II at room temperature with gentle shaking.Completely remove wash buffer in each wash step. Dilute 20X Wash Buffer II with H2O.

Incubation with Biotinylated Antibody Cocktail & Wash

5. Reconstitute the detection antibody by adding 1.4 ml of Sample Diluent to the tube. Spin briefly.

6. Add 80 µl of the detection antibody cocktail to each well. Incubate at room temperature for 1-2 hour.

7. Decant the samples from each well, and wash 5 times (5 mins each) with 150 µl of 1X Wash Buffer I and then 2 times with 150 µl of 1x Wash Buffer II at room temperature with gentle shaking. Completely remove wash buffer in each wash step.

Incubation with Cy3 Equivalent Dye-Streptavidin & Wash

8. After briefly spinning down, add 1.4 ml of Sample Diluent to Cy3 equivalent dye-conjugated streptavidin tube. Mix gently.

9. Add 80 µl of Cy3 equivalent dye-conjugated streptavidin to each well. Cover the device with aluminum foil to avoid exposure to light or incubate in dark room. Incubate at room temperature for 1 hour.

10. Decant the samples from each well, and wash 5 times (5 mins each) with 150 µl of 1X Wash Buffer I at room temperature with gentle shaking. Completely remove wash buffer in each wash step.

Fluorescence Detection

11. Disassemble the device by pushing clips outward from the slide side. Carefully remove the slide from the gasket.

12. Place the slide in the Slide Washer/Dryer (a 4-slide holder/centrifuge tube), add enough 1x

Application Details

	Wash Buffer I (about 30 ml) to cover the whole slide, and then gently shake at room temperature for 15 minutes. Decant Wash Buffer I. Wash with 1x Wash Buffer II (about 30 ml) and gently shake at room temperature for 5 minutes. 13. Remove water droplets completely by gently applying suction with a pipette to remove water droplets. Do not touch the array, only the sides. 14. Imaging: The signals can be visualized through use of a laser scanner equipped with a Cy3 wavelength (green channel) such as Axon GenePix.
Calculation of Results:	Data extraction can be done using the GAL file that is specific for this array along with the microarray analysis software (GenePix, ScanArray Express, ArrayVision, MicroVigene, etc.).
Restrictions:	For Research Use only

Handling

Handling Advice:	Do not touch the surface of the slides, as the microarray slides are very sensitive. Hold the slides by the edges only. Handle all buffers and slides with powder free gloves. Handle glass slide/s in clean environment. The G-Series slides do not have bar codes. To help distinguish one slide from another, transcribe the slide serial number from the slide bag to the back of the slide with a fine point permanent marker. Please write the number on the very bottom edge of the slide, taking care to avoid writing on the array well areas.
Storage:	-20 °C
Storage Comment:	Upon receipt, all components should be stored at -20°C. The kit will retain activity for up to 6 months. Once thawed, the glass slide, standard mix, antibody cocktail and dye-conjugated Streptavidin should be kept at -20°C. All other components may be stored at 4°C. The entire kit should be used within 6 months of purchase.
Expiry Date:	6 months

Publications

Product cited in:	Zheng, Pan, Wang, Liu, Shi, Ding: "HMGB1 Turns Renal Tubular Epithelial Cells into Inflammatory Promoters by Interacting with TLR4 During Sepsis." in: **Journal of interferon & cytokine research : the official journal of the International Society for Interferon and Cytokine Research**, Vol. 36, Issue 1, pp. 9-19, (2016) (PubMed). Chang, Jia, Sun, Xu, Wu, Zhang, Wei: "APRIL promotes proliferation, secretion and invasion of fibroblast-like synoviocyte from rats with adjuvant induced arthritis." in: **Molecular immunology**,

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