

Technical Data Sheet

Mouse Cell Surface Marker Screening Panel

Product Information

Material Number:	562208
Size:	5 tests
Component:	51-9007606AK
Description:	Mouse Cell Surface Marker Screening Panel – Part A
Size:	5 tests (1 ea)
Component:	51-9007606BK
Description:	Mouse Cell Surface Marker Screening Panel – Part B
Size:	5 tests (1 ea)

Description

The BD Lyoplate™ Mouse Cell Surface Marker Screening Panel contains 176 purified monoclonal antibodies to cell surface markers. The panel also contains mouse, rat and hamster immunoglobulin (Ig) isotype controls for assessing nonspecific background staining. The panel can be used for screening cell lines, primary cells or tissue, and is compatible with flow cytometry and bioimaging technology platforms. The panel contains three (3) 96 well plates, each well containing 2.75 µg of antibody, enough for five tests (0.5 µg/test). Biotinylated secondary anti-Ig antibodies and Alexa Fluor® 647 conjugated streptavidin as the tertiary reagent are also included in the panel. This product is compatible with cells expressing fluorescent reporter genes, such as green fluorescent protein (GFP) and can be used with additional antibodies that recognize cell surface and intracellular molecules. Positive hits from screens can be followed-up with either purified or fluorochrome-conjugated antibodies offered by BD Biosciences. To access this content, you can search either the name of the clone and/or the name of the specificity on our website at bdbiosciences.com.

It is important to note the antibodies present in this panel may not recognize all isoforms of each cell surface marker. In addition, antibody clones can behave differently on cell types depending on the availability of epitopes present, i.e., certain epitopes can be occluded by post-translational modifications. Results you obtain in this screen may only be relevant to the antibody clones tested. Moreover, since all the antibodies are provided at the same fixed amounts, they may or may not be at their optimal concentrations. Therefore, it is important to verify positive screening hits with either purified or fluorescent antibodies that are used at optimal concentrations (determined by titration) for result confirmation.

Component 51-9007606AK - Mouse Cell Surface Marker Panel - Part A

Mouse Cell Surface Marker Lyoplate Plate 1 (1 each)

Mouse Cell Surface Marker Lyoplate Plate 2 (1 each)

Mouse Cell Surface Marker Lyoplate Plate 3 (1 each)

Store unopened plates at room temperature (18-25°C).

Antibodies are lyophilized in an aqueous buffered solution containing BSA and ≤ 0.09% sodium azide.

Component 51-9007606BK - Mouse Cell Surface Marker Screening Panel - Part B

Biotin Goat Anti-Rat Ig (1.0 mL)

Biotin Goat Anti-Mouse Ig (0.25 mL)

Biotin Goat Anti-Armenian Hamster Ig (0.25 mL)

Biotin Goat Anti-Syrian Hamster Ig (0.1 mL)

Alexa Fluor® 647 Streptavidin (0.2 mL)

Store the biotinylated secondary antibodies and fluorescent streptavidin protected from light at 4°C.

The biotinylated secondary antibodies are provided in an aqueous buffered solution containing ≤ 0.09% sodium azide.

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Application Notes & Recommended Assay Procedure:

Important instructions before you begin:

- Do not remove the plates from the foil bags until they are ready to be used. The foil bag is the primary moisture barrier. Once the plates are removed from the foil bags, the antibodies must be reconstituted.
- Before removing the foil seal, be sure to centrifuge plates to pellet lyophilized Ab cakes and also use caution when removing foil seal. Please see "Reconstituting the antibody" section below for details.
- After the foil seal is removed and prior to reconstitution, avoid placing the plastic lid or any cover on the plates or resealing the plates with an adhesive-based plate seal. In each case, the resulting static can cause the cakes to dislodge and escape from the wells.
- You may notice that not all lyophilized cakes have the same physical appearance. This is expected and will not affect performance of the antibodies.
- Some cell surface markers are sensitive to enzymatic digestion. When possible use a non-enzymatic cell dissociation buffer for preparing cells for flow cytometry. For enzymatic cell dissociation of cell lines we recommend using Accutase™ (Cat. No. 561527).
- Ensure that cells are in a single cell suspension. A DNase treatment step can mitigate cell clumping.
- Some antibodies to cell surface markers can produce artifacts (false positives and negatives) on fixed cells. If fixation is necessary, staining live cells with subsequent fixation prior to analysis can help reduce these artifacts.

Recommendations for staining with antibodies:

- Evaluating for background staining on cells is recommended by titrating the secondary biotinylated antibodies and Alexa Fluor® 647 Streptavidin as the tertiary reagent before attempting a full screen. Excess secondary antibody and tertiary reagent has been provided. Based on the cell types tested, it is suggested to use the biotinylated anti-mouse, anti-rat and anti-syrian hamster Ig secondary antibodies at 1.25 µg/mL (100 µL per well) and the biotinylated anti-Armenian hamster Ig secondary antibody at 0.6 µg/mL (100 µL per well).
- While the majority of the antibodies in the panel were raised in rat, some of the antibodies were raised in mouse, Syrian hamster or Armenian hamster. To ensure that the appropriate species-specific, biotinylated secondary antibody is used with cells stained with the corresponding primary antibodies, refer to the following Table and to the color-coded plate layout on pages 7-9.

<u>Plate</u>	<u>Secondary</u>	<u>Plate Map Well Color</u>	<u>Wells</u>	<u>Secondary Control Well</u>
1	Rat	Red	All wells	A1
2	Rat	Red	A1 – A12 ; B1 – B12 ; C1 – C8 ; D1 – D7	A1
2	Syrian	Yellow	H1 – H4	H4
3	Armenian	Green	A1 – A12 ; B1 – B12 ; C1 – C2 ; D1 – D6	A1
3	Mouse	Blue	F1 – F12 ; G1 – G10 ; H1 – H6	H6

- Check for any cross-reactivity with the biotinylated secondary antibodies if you plan to treat cells with an activating or inhibitory antibody of your choice (e.g with soluble or plate-bound antibodies).
- This product contains 27 mouse anti-mouse specificity antibodies in Plate 3 (including 5 Ig isotypes). The biotinylated polyclonal goat anti-mouse Ig secondary reagent will bind to cells of the mouse B-cell lineage that express surface Ig. The use of appropriate counterstains to delineate cell types is recommended. For example, an anti-CD3 antibody conjugated to a fluorochrome other than Alexa Fluor® 647 may be used to gate on T cells during analysis.
- Negative control wells containing only biotinylated secondary antibody with Alexa Fluor® 647 Streptavidin and wells with only unstained cells is recommended for each experiment. Well A1 on Plates 1 and 2 can be used for the anti-rat Ig secondary antibody control. Well H4 on Plate 2 can be used for the anti-Syrian hamster Ig secondary antibody control. Well A1 on Plate 3 can be used for the anti-Armenian hamster Ig secondary antibody control. Well H6 on Plate 3 can be used for the anti-mouse Ig secondary antibody control. Any of the remaining buffer wells can be used as unstained cells only control wells.

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- Additional antibodies not currently included on the BD Lyoplate Mouse Cell Surface Screening Panel may be added to the screen in any of the grey wells shown on Plate 2 and 3 maps.
- Mouse BD Fc Block™: Some antibody preparations may bind via the Fc region to Fc receptor bearing cells, resulting in high, non-specific background staining. BD Pharmingen™ Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™) (Cat. No. 553141/553142) may be used to block the Fc-mediated adherence of antibodies to mouse Fc receptors. Since BD Fc Block™ has a rat IgG2b isotype, however, it can only be used with primary antibodies from mouse, Syrian hamster and/or Armenian hamster on Plates 2 and 3. **NOTE: When using BD Fc Block™, secondary anti-Ig antibodies that do not cross-react with its rat IgG2b isotype must be chosen.**
- Armenian hamster Ig isotypes may be used as controls for the 3 antibodies raised from Syrian hamster. For example, Hamster IgG2, λ on Plate 3 (well D4) can be used as an Ig isotype control for the anti-CD28 antibody on Plate 2 (well H1). To use this Ig isotype for the Syrian hamster anti-mouse CD28 antibody, add the anti-Syrian hamster Ig second step along with the anti-Armenian hamster Ig second step in Step 14 described below.
- For flow cytometric analysis, running 500,000 to 1,000,000 cells per well is recommended for best results. However, we have been successful in running as few as 250,000 cells per well.
- For flow cytometric analysis, using a 96-well High Throughput Screening (HTS) Plate Loader is recommended. If a plate loader is not available, transfer stained cells from 96-well plates into Falcon® 12 X 75 mm round bottom tubes (Cat. No. 352008) for manual loading.

Reconstituting the antibody:

- After removing BD Lyoplate™ Mouse Cell Surface Marker Screening Panel plates from foil bags, centrifuge at 300 X g for 5 minutes.
- Hold the plate firmly on the work bench and **gently remove the foil seal** starting from one end and pulling across the plate to completely remove the seal. Once the foil seal is removed, all lyophilized antibodies **must be** immediately reconstituted. Do not replace the lid on the plate prior to reconstitution.
- Using a multi-channel pipette, reconstitute lyophilized antibodies in 110 µl of 1X sterile PBS. This results in an antibody solution that contains five tests (20 µl/test). Be sure to use fresh pipette tips for each row to prevent well-to-well contamination. Allow antibodies to reconstitute for five minutes at room temperature.
- **Store the reconstituted antibodies at 4°C until the cells are prepared for experiments. Reconstituted antibodies can be stored in plates with lids at 4°C for at least 10 days. Seal the plate edges (with lid on) with Parafilm "M"® laboratory film to prevent loss of reconstituted antibody due to evaporation.**

Screening cells by flow cytometry: {~300 mL BD Pharmingen™ Stain Buffer (FBS) is needed for screening in step 6}

1. Prepare a single cell suspension of live cells from a cell line, tissue or a three dimensional culture. For adherent cell lines, using either a mild enzyme such as Accutase™ (Cat. No. 561527) or a non-enzymatic dissociation buffer is recommended.
2. Wash the cells in two to four volumes of 1X PBS. Centrifuge at 300 X g for 5 minutes.
3. Remove any clumps by passing the cells through a Falcon® 40 or 70 µm cell strainer (Corning Cat. No. 352340, Cat. No. 352350).
4. Determine the cell concentration and total number of cells. If you are dissociating tissue or a three dimensional culture, we recommend treating the single cells with DNase to prevent cell clumping. Resuspend cells in the recommended growth media or 1X PBS with calcium and magnesium with the addition of 100 units/mL DNase at 10 million cells per mL. Incubate for 15 minutes at room temperature.
5. Wash the cells in two to four volumes of 1X PBS. Centrifuge at 300 X g for 5 minutes.
6. You will need around 300 mL of BD Pharmingen™ Stain Buffer (FBS) (Cat. No. 554656) for subsequent steps.
7. Resuspend the cells from step 5 in BD Pharmingen™ Stain Buffer. You will need 110 to 220 million cells (in approximately 22 mL total volume) to fill the antibody containing wells and the control wells of the three plates (500,000-1,000,000 cells per well). The minimum number of cells per well will depend on the cytometer and/or loss of cells during washing. We have been successful in running as few as 250,000 cells per well.

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8. Label three Falcon® round bottom 96 well plates (Corning Cat No. 351177) plates 1, 2 and 3 for your sample plates.
9. Using a multi-channel pipette, aliquot 100 µL of cell suspension to required wells of the three labeled round-bottom 96-well plates.
 - a. If you have a limited number of cells, you can omit buffer only control wells from plates 2 and 3. Please refer to the Plate 2 and 3 maps to identify wells that can be excluded, taking into consideration unstained cells and secondary antibody controls.
10. Using a multi-channel pipette, pipette up and down 2-3 times to fully mix the reconstituted antibody from the first row of wells from the BD Lyoplate™ Screening Panel Plate 1. After mixing, add 20 µl of the antibody solution to the cells in the corresponding wells of sample plate 1. Change pipette tips. Continue to add reconstituted antibody to the corresponding sample wells for all remaining wells of each plate. Use fresh tips for every well. Incubate on ice for 20-30 minutes.
11. To wash, add 100 µl of BD Pharmingen Stain Buffer to each well. Centrifuge at 300 X g for 5 minutes.
12. Remove supernatant carefully and wash cells with an additional 200 µl of BD Pharmingen Stain Buffer. Centrifuge at 300 X g for 5 minutes.
13. During the centrifugation step of the final wash, dilute the secondary antibodies in BD Pharmingen Stain Buffer according to the following table:

Secondary Ab	Dilution	Final Concentration (µg/mL)	Volume of Diluted Secondary Ab (mL)
Rat	1:400	1.25	15.0
Syrian	1:400	1.25	0.8
Armenian	1:800	0.60	4.0
Mouse	1:400	1.25	4.0

14. Remove supernatant and apply 100 µl of the appropriate biotinylated secondary antibody directly to cells in each well containing primary antibody as shown in the plate maps (pages 7-9). Also, select an additional well as a secondary plus tertiary reagent control for each of the 4 secondary antibodies. Use the table below for reference. Use remaining wells in sample plate 2 or 3 that do not contain antibody (grey colored plate map wells) to setup unstained cells only controls.

Plate	Secondary	Plate Map Well Color	Wells	Secondary Control Well
1	Rat	Red	All wells	A1
2	Rat	Red	A1 – A12 ; B1 – B12 ; C1 – C8 ; D1 – D7	A1
2	Syrian	Yellow	H1 – H4	H4
3	Armenian	Green	A1 – A12 ; B1 – B12 ; C1 – C2 ; D1 – D6*	A1
3	Mouse	Blue	F1 – F12 ; G1 – G10 ; H1 – H6	H6

* If the use of Armenian hamster Ig isotype controls for the 3 Syrian hamster antibodies is desired, add the biotinylated anti-Syrian hamster secondary antibody along with the biotinylated anti-Armenian hamster secondary antibody to wells D1 (Arm IgG1, κ), D3 (Arm IgG2, κ) and D4 (Arm IgG2, λ).

15. Incubate for 20-30 minutes on ice in the dark.
16. To wash, add 100 µl of BD Pharmingen Stain Buffer to each well. Centrifuge at 300 X g for 5 minutes.
17. Remove supernatant and wash cells with an additional 200 µL of BD Pharmingen Stain Buffer. Centrifuge at 300 x g for 5 minutes.
18. Remove supernatant and add 100 µL of Alexa Fluor® 647 Streptavidin to all wells containing cells stained with the biotinylated secondary antibodies (not to wells selected as unstained cell controls). Dilute the Alexa Fluor® 647 Streptavidin 1:4000 (0.5 µg/mL) in 22 mL of BD Pharmingen Stain Buffer.
19. Incubate for 20-30 minutes on ice in the dark.
20. To wash, add 100 µl of BD Pharmingen Stain Buffer to each well. Centrifuge at 300 X g for 5 minutes.

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21. Remove supernatant and wash cells with an additional 200 μ L of BD Pharmingen Stain Buffer. Centrifuge at 300 x g for 5 minutes.
22. At this point you may wish to fix your cells prior to analysis. To fix, remove supernatant and add 100 μ L of 4% paraformaldehyde in 1X PBS or BD Cytofix™ Fixation Buffer (Cat. No. 554655) per well and incubate for 10 minutes. If you do not wish to fix your cells go to step 24.
23. Wash cells twice with 1X PBS. Centrifuge at 300 X g for 5 minutes.
24. Remove supernatant and resuspend cells in 150 μ L of BD Pharmingen Stain Buffer per well.
25. Analyze your samples on a flow cytometer. We recommend collecting at least 10,000 events per well. While the first plate is being read, store the other plates on ice in the dark.

Screening cells by bioimaging:

1. Seed the cells in appropriate culture medium at an appropriate cell density in a Falcon® 96-well Imaging Plate (Cat. No. 353219), and culture cells to an appropriate density. We recommend 70-80% confluence for imaging screens.
2. Cell surface staining with antibodies from the BD Lyoplate should be performed on live cells, as cellular fixation can cause artifacts (false positive and/or negative signals) with some cell surface markers. In cases where cells must be fixed prior to staining, we recommend confirming any positive hits with a live sample stain using imaging or flow cytometry.
3. Using a multi-channel pipette add 20 μ L of each reconstituted antibody to the corresponding wells of your sample plates and incubate on ice for 20-30 minutes. Stain cells directly in 50 to 100 μ L of fresh growth media. If staining fixed cells, stain cells in 1X PBS.
4. Wash cells twice in 100 μ L 1X PBS.
5. Dilute secondary antibodies in growth media according to the following table:

<u>Secondary Ab</u>	<u>Dilution</u>	<u>Final Concentration (μg/mL)</u>	<u>Volume of Diluted Secondary Ab (mL)</u>
Rat	1:400	1.25	15.0
Syrian	1:400	1.25	0.8
Armenian	1:800	0.60	4.0
Mouse	1:400	1.25	4.0

6. Remove supernatant and apply 100 μ L of the appropriate biotinylated secondary antibody directly to each well containing cells stained with the primary antibody as shown in the plate map. Also, select an additional well as a Secondary plus Tertiary Reagent control for each of the 4 secondary antibodies. Use the table below for reference. Use remaining wells in sample plate 3 that do not contain antibody (grey colored plate map wells) to setup unstained cell controls.

<u>Plate</u>	<u>Secondary</u>	<u>Plate Map Well Color</u>	<u>Wells</u>	<u>Secondary Control Well</u>
1	Rat	Red	All wells	A1
2	Rat	Red	A1 – A12 ; B1 – B12 ; C1 – C8 ; D1 – D7	A1
2	Syrian	Yellow	H1 – H4	H4
3	Armenian	Green	A1 – A12 ; B1 – B12 ; C1 – C2 ; D1 – D6	A1
3	Mouse	Blue	F1 – F12 ; G1 – G10 ; H1 – H6	H6

7. Incubate for 20-30 minutes on ice in the dark.
8. Remove supernatant and wash cells twice in 100 μ L 1X PBS.
9. Dilute the Alexa Fluor® 647 Streptavidin 1:4000 (0.5 μ g/mL) in 22 mL of growth media. Add 100 μ L of Alexa Fluor® 647 Streptavidin to all wells containing cells stained with the biotinylated secondary antibodies (not to the wells selected as unstained cell controls).

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10. Incubate for 20-30 minutes on ice in the dark.
11. Remove supernatant and wash cells twice in 100 μ L 1X PBS.
12. At this point you may wish to fix your cells prior to analysis. To fix, remove supernatant and add 100 μ L of 4% paraformaldehyde in 1X PBS or BD Cytotfix Fixation Buffer per well and incubate for 10 minutes. If you do not wish to fix your cells go to step 14.
13. Remove the fixative from the wells, and wash the wells twice with 100 μ L of 1X PBS.
14. Add 100 μ L 1X PBS with a cell-permeable nucleic acid stain, such as Hoechst 33342 Solution (Cat. No. 561908).
15. Analyze your samples on a high content bioimager.

Suggested Companion Products

Description	Size	Catalog Number
BD Pharmingen™ Stain Buffer (FBS)	500 mL	554656
BD Cytotfix™ Fixation Buffer	100 mL	554655
BD Accutase™	100 mL	561527
BD Pharmingen™ Hoechst 33342 Solution	1 mg/mL	561908
BD Pharmingen™ Fc Block	0.5 mg/mL	553141 or 553142

Related Products

Description	Size	Catalog number
Falcon® 96-well Microplates, Black/Clear With Lid, for High-Content Imaging Assays	32/case	353219
Falcon® 96-well Microplates, Round Bottom with Lid, for Flow Cytometry Analysis	50/case	351177
Falcon® Round Bottom Tube, 12 x 75 mm	1000/case	352008

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4. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
5. This product may be covered by US Patent No. 5,543,320.
6. US Patent No. 5,994,515, University of Pennsylvania.
7. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
8. 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.

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	Plate 1	2	3	4	5	6	7	8	9	10	11	12
A	Buffer	CD2	CD4	CD5 (Ly-1)	CD8a (Ly-2)	CD9	CD11a	CD11b	CD13	CD14	CD16/CD32	CD18
B	CD19	CD21/CD35	CD23	CD24	CD25	CD26	CD29	CD31	CD34	CD35	CD38	CD41
C	CD43	CD44	CD45	CD45R	CD45RA	CD45RC	CD47	CD49b	CD49d	CD49e	CD51	CD53
D	CD62E	CD62L	CD70	CD71	CD72b/c	CD73	CD80	CD83	CD86	CD90.2	CD94	CD98
E	CD102	CD103	CD104	CD106	CD117	CD121a	CD121b	CD122	CD123	CD124	CD125	CD126
F	CD127	CD131	CD132	CD134	CD135	CD137	CD138	CD140a	CD144	CD147	CD153	CD162
G	CD172a	CD179a	CD179b	CD180	CD184 (CXCR4)	CD185 (CXCR5)	CD195	CD197	CD200	CD209a	CD210	CD223
H	CD244.1	CD252	CD253	CD254 (RANKL)	CD267	CD273	CD274	CD278	CD284	CD309 (Flk-1, VEGF-R2)	CD314	CD326

Plate 1											
Specificity	Alternate Names			Clone	Isotype	Specificity	Alternate Names			Clone	Isotype
CD2	LFA-2, Ly-37, Ly37			RM2-5	Rt IgG2b,λ	CD102	ICAM-2, Inter cellular adhesion molecule 2, Ly-60			3C4(MIC2/4)	Rt IgG2a,κ
CD4	L3T4, Ly-4			GK1.5	Rt IgG2b,κ	CD103	Itgae, Integrin alpha-e, Integrin alpha IEL			M290	Rt IgG2a,κ
CD5	Ly-1, Lyt-1, Ly-12, Ly-A			53-7.3	Rt IgG2a,κ	CD104	Itgb4, Integrin beta-4			346-11A	Rt IgG2a,κ
CD8A	Ly-2, Lyt-2, Ly-B, Ly-35, Ly-B			53-6.7	Rt IgG2a,κ	CD106	Vcam1, Vcam-1, Vascular cell adhesion molecule 1			429(MVCAM.A)	Rt IgG2a,κ
CD9	Tspan29, Tetraspanin 29			KMC8	Rt IgG2a,κ	CD117	Kit, c-kit, SCFR, Stem cell growth factor Receptor, Steel factor Receptor			2B8	Rt IgG2b,κ
CD11a	Itgal, Integrin alpha L, Ly-15, Ly-21, LFA-1a			M17/4	Rt IgG2a,κ	CD121a	I			35F5	Rt IgG1,κ
CD11b	Itgam, Integrin alpha M, Ly-40, CR3a, Mac-1a			M1/70	Rt IgG2b,κ	CD121b	IIIR2, IL-1R2, IL-1RII, IL-1R beta, IL-1RB, IL-1 Receptor Type II			4E2	Rt IgG2a
CD13	Anpep, Apn, Lap-1, Aminopeptidase N, gp150			R3-242	Rt IgG1,κ	CD122	IL2rb, IL-2RB, IL-2/15 Receptor beta, IL-2 and IL-15 Receptor beta			TM-BETA1	Rt IgG2b
CD14	Mo2, LPS Receptor			RMC5-3	Rt IgG1,κ	CD123	II3ra, IL-3R alpha, IL-3RA, IL-3 Receptor alpha			5B11	Rt IgG2a,κ
CD16/CD32	Fcgr3, FcgammaRIII/Fcgr2, FcgammaRII, Ly-17			2.4G2	Rt IgG2b,κ	CD124	II4ra, IL-4R alpha, IL-4RA, IL-4 Receptor alpha			ML4R-M1	Rt IgG2a,κ
CD18	Itgb2, Integrin beta 2, LFA-1/Mac-1/CR3 beta			C71/16	Rt IgG2a,κ	CD125	II5ra, IL-5R alpha, IL-5RA, IL-5 Receptor alpha			T21	Rt IgG1,λ
CD19	B4; B-lymphocyte antigen CD19			1D3	Rt IgG2a,κ	CD126	II6ra, IL-6R alpha, IL-6RA, IL-6 Receptor alpha			D7715A7	Rt IgG2b,κ
CD21/CD35	CR2/CR1			7G6	Rt IgG2b,κ	CD127	II7r, IL-7R alpha, IL-7RA, IL-7 Receptor alpha			B12-1	Rt IgG2a,λ
CD23	Fcr2, FceRII, FcεpsilonRII, Ly-42			B3B4	Rt IgG2a,κ	CD131	Csf2rb/Csf2rb2, AIC2B/AIC2A, bc/bIL3, II3rb1/II3rb2			JORO50	Rt IgG1,κ
CD24	Heat Stable Antigen, HSA, Ly-52, Nectadrin			M1/69	Rt IgG2b,κ	CD132	II2rg, IL-2 Receptor gamma, Cytokine Receptor Common gamma			TUGM2	Rt IgG2b,κ
CD25	II2ra, IL-2 Receptor alpha, IL-2R alpha, Ly-43, p55			PC61	Rt IgG1,λ	CD134	Tnfrsf4, Ly-70, OX-40, OX40L Receptor, ACT35 antigen			OX-86	Rt IgG1,κ
CD26	Dpp4, Dipeptidyl peptidase 4, DPP IV, THAM			H194-112	Rt IgG2a,κ	CD135	Flk3, Fms-like tyrosine kinase 3, Flk-2, Ly-72			A2F10.1	Rt IgG2a,κ
CD29	Itgb1, Integrin beta-1, VLA-4 beta, gpIIa			9EG7	Rt IgG2a,κ	CD137	Tnfrsf9, 4-1BB, Ly-63, ILA			1AH2	Rt IgG1,κ
CD31	Pecam1, endoCAM, platelet endothelial cell adhesion molecule			MEC13.3	Rt IgG2a,κ	CD138	Sdc1, Syndecan-1, Synd1, Syn-1, synstatin, Sstn			2B1-2	Rt IgG2a,κ
CD34	Mucosialin			RAM34	Rt IgG2a,κ	CD140a	Pdgrfa, PDGF Receptor alpha, PDGF-R alpha			APAS5	Rt IgG2a,κ
CD35	CR1, Complement Receptor 1, C3b Receptor			8C12	Rt IgG2a,κ	CD144	Cdh5, Cadherin 5, VE-Cadherin, 7B4, VECD			11D4.1	Rt IgG2a,κ
CD38	ADP-ribosyl cyclase 1, T10, Cyclic ADP-ribose hydrolase 1			90	Rt IgG2a,κ	CD147	Bsg, Basigin, HT7, gp42, Neurothelin			RL73	Rt IgG2a,κ
CD41	Itga2b, Integrin alpha-2b, GPIIb, Platelet membrane glycoprotein IIb			MWREG30	Rt IgG1,κ	CD153	Tnfrsf8, CD30 Ligand, CD30L, CD30LG			RM153	Rt IgG2b
CD43	Sps, Sialophorin, Leukosialin, Galgp, Ly-48			S7	Rt IgG2a,κ	CD162	Selplg, P-selectin glycoprotein ligand 1, PSGL-1			2PH1	Rt IgG1,κ
CD44	Ly-24, Pgp-1, ECMRIII, HUTCH-1, Hermes, Hyaluronate Receptor			IM7	Rt IgG2b,κ	CD172a	Sirpa, SHPS-1, BIT, P84 Antigen, SIRP, SHP-1, Ptpns1			P84	Rt IgG1,κ
CD45	Ptpcr, Leukocyte Common Antigen, LCA			30-F11	Rt IgG2b,κ	CD179a	Vpreb1, Pre-B lymphocyte gene 1, Immunoglobulin iota chain			R3/VPREB	Rt IgG2a,κ
CD45R	Ptpcr, B220, Ly-5, Lyt-4			RA3-6B2	Rt IgG2a,κ	CD179b	Vpreb2, Igll1, Igl-5, Ig lambda-5			LM34	Rt IgG2a,κ
CD45RA	Ptpcr, Ly-5, Lyt-4			1A.8	Rt IgG2b,κ	CD180	Ly-78, RP105			RP14	Rt IgG2a,κ
CD45RC	Ptpcr, Ly-5, Lyt-4			DNL-1.9	Rt IgG2a,κ	CD184 (CXCR4)	CKR			2B11/CXCR4	Rt IgG2b
CD47	Itgp, integrin-associated protein, IAP			MIAP301	Rt IgG2a,κ	CD185 (CXCR5)	CXCR5, BLR1, Gpcr6, MDR15			2G8	Rt IgG2a
CD49b	Itga2, Integrin alpha-2, DX5, VLA-2a, GPIa			DX5	Rt IgM,κ	CD195	CCR5, Cmkbr5, AM4-7, MIP-1 alpha Receptor			C34-3448	Rt IgG2c,κ
CD49d	Itga4, Integrin alpha-4, VLA-4a, LPAM alpha			9C10(MRF4.B)	Rt IgG2a,κ	CD197	CCR7, EBI-1, BLR2, CMKBR7, MIP-3 beta Receptor			4B12/CCR7.1	Rt IgG2a
CD49e	Itga5, Integrin alpha-5, VLA-5a, Fnra, Fibronectin Receptor alpha			5H10-27	Rt IgG2a,κ	CD200	OX-2, Mox2			OX-90	Rt IgG2a,κ
CD51	Itgav, Integrin alpha-v, VNRA, Vitronectin Receptor alpha			RMV-7	Rt IgG1,κ	CD209a	DC-SIGN, CDSIGN, CIRE, CLEC4L			5H10/CIRE	Rt IgG2a
CD53	OX-44, TSPAN25, Tetraspanin-25			OX-79	Rt IgM,κ	CD210	II10ra, IL-10 Receptor alpha, IL-10Ra, IL-10R1			1B1.3A	Rt IgG1,κ
CD62E	Sele, E-Selectin, ELAM-1, LECAM-2			10E9.6	Rt IgG2a,κ	CD223	LAG3, LAG-3, Lymphocyte activation gene 3, FDC protein, Ly-66			C9B7W	Rt IgG1,κ
CD62L	Sell, L-Selectin, LECAM-1, Lnhp, Ly-22, Ly-m22, Lyam-1, Lyam1			MEL-14	Rt IgG2a,κ	CD244.1	2B4, C9.1, Ly90, NAIL, Nmrk, NKR2B4, SLAMF4			C9.1	Rt IgG2b,κ
CD70	CD27 Ligand, CD27L, Tnfsf7			FR70	Rt IgG2b,κ	CD252	Tnfsf4, OX-40 Ligand, OX40L, gp34, Txgp1l, Ath1, CD134L			RM134L	Rt IgG2b,κ
CD71	Tfrc, Transferrin Receptor, Mtrv1, TFR, TR, Trfr, TFR1			C2	Rt IgG1,κ	CD253	Tnfsf10, TRAIL, APO-2L, TL2, Ly81, Trail, APO-2L			N2B2	Rt IgG2a,κ
CD72 b/c AlloA	Lyb-2, Ly-19, Ly-m19, Ly-32, Ly-32			JY/93	Rt IgG1,κ	CD254 (RANKL)	Tnfsf11, ODF, OPG, OPGL, RANKL, Trance, SODF			1K22-5	Rt IgG2a,κ
CD73	NT5e, NT, N5e, Nte, Ecto-5'-nucleotidase			TY/23	Rt IgG2a,κ	CD267	Tnfrsf13b, TACI			8F10	Rt IgG2a,κ
CD80	B7/BB1, B7-1, Ly-53			1G10/B7	Rt IgG2a,κ	CD273	PdcclIg2, Programmed cell death 1 ligand 2, PD-L2, B7-DC, Btdc			TY25	Rt IgG2a,κ
CD83	HB15			MICHEL-19	Rt IgG1,κ	CD274	Programmed cell death 1 ligand 1, PD-L1, Pdccl1l1, Pdccl1g1, B7-H1			MIH5	Rt IgG2a,λ
CD86	B7-2, B70, Ly-58, ETC-1			PO3	Rt IgG2b,κ	CD278	ICOS, Inducible T-cell co-stimulator, Ly115, H4, AILIM, CCLP, CRP-1			7E.17G9	Rt IgG2b,κ
CD90.2	Thy1, Thy-1.2, q-C3H			30-H12	Rt IgG2b,κ	CD284	TLR4, Toll-like receptor 4, Ly87, Ran/M1, Rasi2-8, Lps			MTS510	Rt IgG2a,κ
CD94	Klrtd1, Killer cell lectin-like Receptor subfamily D member 1, KP43			18D3	Rt IgG2a,κ	CD309	KDR, VEGFR2, Flk-1			Avas 12o1	Rt IgG2a,κ
CD98	Slc3a2, 4F2HC, Ly-10, Mgp-2hc, Mdu1, NACAE			H202-141	Rt IgG2a,κ	CD314	KLRK1, NKG2D, KLR			CX5	Rt IgG1,κ
						CD326	Epcam, epithelial cell adhesion molecule, EGP314, Ly-74, Tacstd1, Trool			G8.8	Rt IgG2a,κ

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Plate 2	2	3	4	5	6	7	8	9	10	11	12	
A	Buffer	CD335 (NKP46)	4-1BB Ligand (CD137L)	Crry/p65	Dendritic Cells	Early B Lineage	F4/80-like receptor	GITR	I-A/I-E	IL-21 Receptor	Integrin β7 chain	LPAM-1
B	Ly-6A/E (Sca-1)	Ly-6D (ThB)	Ly-6G	Ly-6G Ly-6C	CD107b	MAc-CAM-1	MD-1 (Ly-86)	NKG2 A/C/E	NK-T/NK Cell Antigen	Panendothelial Cell Antigen	PIR-A/B	Pre BCR
C	Siglec-F (Siglec5)	Syndecan 4 (Sdc4)	T/B cell activation antigen	TER-119 (Erythroid Cells)	Ig λ_{123} light chain	IgD	IgE	IgM	Buffer	Buffer	Buffer	Buffer
D	Rat IgG1, κ	Rat IgG1, λ	Rat IgG2a, κ	Rat IgG2a, λ	Rat IgG2b, κ	Rat IgG2c, κ	Rat IgM, κ	Buffer	Buffer	Buffer	Buffer	Buffer
E	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer
F	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer
G	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer
H	CD28	KLRG1	V γ 3 TCR	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer

Plate 2			
Specificity	Alternate Names	Clone	Isotype
CD335 (Nkp46)	Ncr1, Natural cytotoxicity triggering receptor 1, Ly-94, MAR1	29A1.4	Rt IgG2a, κ
4-1BB Ligand	Tnfsf9, CD137L, Ly63l	TKS-1	Rt IgG2a, κ
Crry/p65	Cr1l, Complement component (3b/4b) receptor 1-like, Mcl, mCRY, Cry	1F2	Rt IgG2a, κ
Dendritic Cells	DC specific marker 33D1	33D1	Rt IgG2b, κ
Early B Lineage		493	Rt IgG2a, κ
F4/80-like Receptor	Emr4, EGF-like Module Receptor 4, Egf-tm7, Fire, Gpr127	6F12	Rt IgG2a, κ
GITR	Tnfrsf18, CD357, Glucocorticoid-induced TNFR-related Protein, AITR	DTA-1	Rt IgG2b, λ
I-A/I-E	H-2 Class II Histocompatibility antigen I-A/I-E	2G9	Rt IgG2a, κ
IL-21 Receptor	Il21r, CD360, IL-21R, Interleukin 21 receptor, NR8, LR-beta	4A9	Rt IgG2a, κ
Integrin β7 chain	Itgb7, Ly69, Integrin beta 7, Integrin beta-P, M290 IEL antigen	FIB27	Rt IgG2a, κ
LPAM-1	α4β7, Integrin alpha 4/beta 7, CD49d/Integrin beta 7	DATK32	Rt IgG2, κ
Ly-6A/E	Ly6a, Lymphocyte Antigen-6A/E, Sca-1, Sca1, Stem Cell Antigen 1, TAP	E13-161.7	Rt IgG2a, κ
Ly-6D	Ly6d, Lymphocyte Antigen-6D, Ly-61, Ly61, ThB, Thymocyte B Cell Ag	49-H4	Rt IgG2c, κ
Ly-6G	Ly6g, Lymphocyte Antigen-6G, Gr-1, Gr1	1A8	Rt IgG2a, κ
Ly-6G/Ly-6C	Ly6g/Ly6C, Lymphocyte Antigen-6G/6C	RB6-8C5	Rt IgG2b, κ
CD107b	Lamp2, Lysosomal-associated Membrane Protein 2, CD107b, Mac3	M3/84	Rt IgG1, κ
MAcCAM-1	Madcam1, Mucosal vascular addressin cell adhesion molecule 1	MECA-89	Rt IgG2a, κ
MD-1	Ly86, Ly-86, Lymphocyte antigen-86, MD1, MMD-1	MD14	Rt IgG2a, κ
NKG2A/C/E	Klrc1/2/3, Killer cell lectin-like receptor subfamily C, members 1/2/3	20D5	Rt IgG2a, κ
NKT/NK Cell Antigen	Icam1, CD54, ICAM-1, Intercellular adhesion molecule 1, Ly-47	USA2-13	Rt IgG2a, κ
Panendothelial Cell Ag	Plvap; Pv1; MECA32; Plasmalemma vesicle-associated protein	MECA-32	Rt IgG2a, κ
PIR-A/B	Paired immunoglobulin-like receptors-A/B	6C1	Rt IgG1, κ
Pre-BCR	Pre-B Cell Receptor	SL156	Rt IgG2a, κ
Siglec-F	Siglec5, Sialic acid binding Ig-like lectin 5, CD170	E50-2440	Rt IgG2a, κ
Syndecan-4	Sdc4, Ryudocan core protein	KY/8.2	Rt IgG2a, κ
T/B Cell Activation Ag		GL7	Rt IgM, κ
Erythroid Cells	TER-119, Ly-76	TER-119	Rt IgG2b, κ
Ig $\lambda_{1,2,3}$ Light Chain	Igcl1/2/3, Immunoglobulin lambda constant 1/2/3	R26-46	Rt IgG2a, κ
IgD	IGHD, Igh-5, Immunoglobulin heavy constant delta	11-26C.2A	Rt IgG2a, κ
IgE	Igh-7, Immunoglobulin heavy chain 7, Heavy chain of IgE	R35-72	Rt IgG1, κ
IgM	Ighm, Immunoglobulin heavy constant mu, Igh6	R6-60.2	Rt IgG2a, κ
Rat IgG1, κ IC	Rat IgG1, κ Isotype Control	R3-34	Rt IgG1, κ
Rat IgG1, λ IC	Rat IgG1, λ Isotype Control	A110-1	Rt IgG1, λ
Rat IgG2a, κ IC	Rat IgG2a, κ Isotype Control	R35-95	Rt IgG2a, κ
Rat IgG2a, λ IC	Rat IgG2a, λ Isotype Control	B39-4	Rt IgG2a, λ
Rat IgG2b, κ IC	Rat IgG2b, κ Isotype Control	A95-1	Rt IgG2b, κ
Rat IgG2c, κ IC	Rat IgG2c, κ Isotype Control	A23-1	Rt IgG2c, κ
Rat IgM, κ IC	Rat IgM, κ Isotype Control	R4-22	Rt IgM, κ
CD28	Cd28, T-cell-specific surface glycoprotein CD28	37.51	Syr IgG2, λ 1
KLRG1	Klrg1, MAFA, 2F1-Ag	2F1	Syr IgG2, κ
V γ 3 TCR	Tcrg3, Tcrg T-cell receptor gamma chain 3	536	Syr IgG1, κ

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	Plate 3	2	3	4	5	6	7	8	9	10	11	12
A	Buffer	CD3e	CD11c	CD27	CD30	CD40	CD42d	CD48	CD54	CD55	CD61	CD69
B	CD79b	CD81	CD95	CD119	CD120a	CD120b	CD152	CD154	CD279	γδ-TCR	gp49 R	H2-M3
C	IFN-γ Receptor β chain	TCR β chain	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer
D	Ham IgG1, κ	Ham IgG1, λ	Ham IgG2, κ	Ham IgG2, λ	Ham IgG3, λ	Ham IgM, λ	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer
E	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer
F	CD22.2	CD45.1	CD45.2	CD64 a/b	CD72 a/b/d	CD157	CD212	CD244.2	H-2D ^b	H-2K ^b	H-2K ^d	H-2K ^k
G	H-2K ^q	H-2K ^s	IFN-α/β Receptor1	Ly-49C Ly-49I	Ly-51	NK-1.1	Pre TCR α chain	QA-1B	SSEA-1	SSEA-4	Buffer	Buffer
H	Mouse IgG1, κ	Mouse IgG2a, κ	Mouse IgG2b, κ	Mouse IgG3, κ	Mouse IgM, κ	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer	Buffer

Plate 3																			
Specificity	Alternate Names				Clone	Isotype	Specificity	Alternate Names				Clone	Isotype						
CD3e	CD3 epsilon, CD3, T3/Leu4 epsilon				145-2C11	Arm IgG1,κ	CD22.2	Lyb-8.2, Siglec-2, Siglec2, B-cell receptor CD22				CY34.1	Ms IgG1,κ						
CD11c	Itgax, Integrin alpha-X, CR4, Complement receptor-4				HL3	Arm IgG1,λ2	CD45.1	PTPRCa, Ly-5.1, Ly-5a				A20	Ms IgG2a,κ						
CD27	T14, s152, Tnfrs7, Tp55				LG.3A10	Arm IgG1,κ	CD45.2	PTPRCb, Ly-5.2, Ly-5b				104	Ms IgG2a,κ						
CD30	Tnfrs8, Ki, Ki-1				MCD30.1	Arm IgG1,κ	CD64 a/b AlloAgs	Fcgr1, FcγγmaR1, Fc-gamma receptor 1				X54-5/7.1	Ms IgG1,κ						
CD40	gp39 receptor, Tnfrs5, Bp50				HM40-3	Arm IgM,κ	CD72 a/b/d AlloAgs	Lyb-2, Ly-m19, Ly-32				K10.6	Ms IgG2b,κ						
CD42d	Gp5, GPV, Glycoprotein 5, Platelet glycoprotein V				1C2	Arm IgG3,λ1	CD157	Bst1, Ly-65, BP-3, Bone marrow stromal cell antigen 1				BP-3	Ms IgG2b,κ						
CD48	Blast-1, Hulym3, BCM1, OX-45, MEM-102, SLAMF2, Sgp-60				HM48-1	Arm IgG1,λ3	CD212	II12rb1, IL-12R-beta-1, IL-12beta1, IL-12beta, CD212b1				114	Ms IgG2a,κ						
CD54	Icam1, ICAM-1, Ly-47, MALA-2, myD10				3E2	Arm IgG1,κ	CD244.2	Cd244, CD244 2B4, Ly90, NAIL, Nmrk, NKR2B4, SLAMF4				2B4	Ms IgG2b,κ						
CD55	Complement decay accelerating factor GPI-anchored, Daf, Daf-GPI, Daf1, GPI-DAF				RIKO-5	Arm IgG3,λ1	H-2D ^b	H-2Db MHC class I alloantigen				KH95	Ms IgG2b,κ						
CD61 (Integrin β3)	Itgb3, Integrin beta 3, GP3A, Platelet membrane glycoprotein IIIa				2C9.G2	Arm IgG1,κ	H-2K ^b	H-2Kb MHC class I alloantigen				AF6-88.5	Ms IgG2a,κ						
CD69	AIM, Very Early Activation Antigen, VEA				H1.2F3	Arm IgG1,λ3	H-2K ^d	H-2Kd MHC class I alloantigen				SF1-1.1	Ms IgG2a,κ						
CD79b	Igb, Ig beta, B29, BCR complex-associated protein beta chain				HM79B	Arm IgG2,λ1	H-2K ^k	H-2Kk MHC class I alloantigen				AF3-12.1	Ms IgG1,κ						
CD81	Tapa1, TAPA-1, Tspan28				EAT2	Arm IgG,κ	H-2K ^q	H-2Kq MHC class I alloantigen				KH114	Ms IgG2a,κ						
CD95 (Fas)	Fas, APO-1, APO1, APT1, TNFR6, Tnfrs6, lpr				JO2	Arm IgG2,λ2	H-2K ^s	H-2Ks MHC class I alloantigen				KH49	Ms IgM,κ						
CD119 (IFNγR1)	Ifngr1, IFN-gamma Receptor alpha, IFNγR alpha, Ifngra				2E2	Arm IgG1,κ	IFN-α/β receptor 1	Ifnar1, Ifar, Ifrc, CD118				MAR1-5A3	Ms IgG1,κ						
CD120a	Tnfrsf1a, TNFR1, TNF-R1, TNFR-R1, TNFRp55, TNF Receptor Type I				55R-286	Arm IgG1,κ	LY-49C LY-49I	Klra3/Klra9, SE6, Nk2.1				SE6	Ms IgG2a,κ						
CD120b	Tnfrsf1b, TNFR2, TNF-R2, TNF-R1I, TNFRp75, TNF Receptor Type II				TR75-89	Arm IgG1,λ3	LY-51	Enpep, glutamyl aminopeptidase, APA, Bp-1/6C3				BP-1	Ms IgG2a,κ						
CD152	Ctla4, CTLA-4, Cytotoxic T-lymphocyte-associated protein 4, Ly-56				UC10-4F10	Arm IgG1,κ	NK-1.1	Klrb1c, Ly59, CD161, Ly55c, NKR1				PK136	Ms IgG2a,κ						
CD154	Cd40lg, Tnfsf5, gp39, CD40 Ligand, CD40L, Ly-62, HIGM1, T-BAM, TRAP				MR1	Arm IgG3,κ	Pre-TCR α chain	Ptcrα, pT-alpha				2F5	Ms IgG1,κ						
CD279	Pdcd1, Pdc1, Pd1, PD-1, Programmed death-1, Ly101				J43	Arm IgG2,κ	QA-1B	H2-T23, histocompatibility 2 T region locus 23, 37b, 37c, Qa-1, T18c				6A8.6F10.1A6	Ms IgG1,κ						
γδ-TCR	T-cell receptor gamma delta				GL3	Arm IgG2,κ	SSEA-1	Fut4, 3-FAL, LeX antigen, CD15				MC480	Ms IgM,κ						
gp49 Receptor	Ulrb4, Leukocyte immunoglobulin-like receptor subfamily B, CD85K, ILT3				H1.1	Arm IgG3,κ	SSEA-4	Stage specific embryonic antigen 4				MC813-70	Ms IgG3,κ						
H2-M3	H-2M3, Histocompatibility 2 M region locus 3, MHC class I-b antigen M3				130	Arm IgG1,κ	Ms IgG1, κ IC	Mouse IgG1,κ Isotype Control				MOPC-31C	Ms IgG1,κ						
IFN-γ R β Chain	Ifngr1, IFN-gamma Receptor alpha, IFNγR alpha, Ifngra				MOB-47	Arm IgG	Ms IgG2a, κ IC	Mouse IgG2a,κ Isotype Control				G155-178	Ms IgG2a,κ						
TCR β chain	Tcrb, T-cell receptor beta chain, Tib, TCRbeta				H57-597	Arm IgG2,λ1	Ms IgG2b, κ IC	Mouse IgG2b,κ Isotype Control				MPC-11	Ms IgG2b,κ						
Ham IgG1,κ IC	Hamster IgG1,κ Isotype Control				A19-3	Arm IgG1,κ	Ms IgG3, κ IC	Mouse IgG3,κ Isotype Control				A112-3	Ms IgG3,κ						
Ham IgG1,λ IC	Hamster IgG1,λ Isotype Control				G235-2356	Arm IgG1,λ	Ms IgM, κ IC	Mouse IgM,κ Isotype Control				G155-228	Ms IgM,κ						
Ham IgG2,κ IC	Hamster IgG2,κ Isotype Control				B81-3	Arm IgG2,κ													
Ham IgG2,λ IC	Hamster IgG2,λ Isotype Control				Ha4/8	Arm IgG2,λ													
Ham IgG3,λ IC	Hamster IgG3,λ Isotype Control				A19-4	Arm IgG3,λ													
Ham IgM,λ IC	Hamster IgM,λ Isotype Control				G235-1	Arm IgM													

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