



Enabling Legendary Discovery™

LEGENDScreen™

Lyophilized Antibody Array

Catalog Number: 700009

Size: 1 kit, ready to use
1 test per antibody/well

Reactivity: Mouse

Antibody Format: PE-conjugated, lyophilized

Configuration: 264 pre-titrated antibodies, including 253 specificities and 11 isotype controls, arrayed on three 96-well plates, with one specificity per well

It is highly recommended that this manual be read in its entirety before using this product.

Do not use this kit beyond the expiration date

For research use only

BioLegend, Inc
Biolegend.com

For Research Purposes Only. Not for use in diagnostic or therapeutic procedures. Purchase does not include or carry the right to resell or transfer this product either as a stand-alone product or as a component of another product. Any use of this product other than the permitted use without the express written authorization of BioLegend is strictly prohibited.

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Description

The LEGENDScreen™ Mouse (PE) kit contains 253 PE-conjugated monoclonal antibodies to cell surface markers as well as 11 mouse, rat, or hamster Ig isotype controls in a convenient 96 well, ready to use format. The kit can be used for screening cell lines, primary cells such as splenocytes, bone marrow-derived cells and cells isolated from tissues. If other co-stains are needed, they can be added to the wells following reconstitution. Positive “hits” from the screening can be quickly identified based on the plate map and the catalog number and clone information can be obtained using the tables on pages 14 through 22. Analysis with individual fluorochrome-conjugated antibodies should be performed to confirm the screening results.

The LEGENDScreen™ Mouse (PE) Kit provides a convenient, easy to use, and powerful tool for immunology, stem cell, and cancer research.

Materials Provided

Description	Quantity	Size	Part #
Mouse Cell Screening (PE) Kit Plate #1	1 plate	96-well	750002444
Mouse Cell Screening (PE) Kit Plate #2	1 plate	96 well	750002445
Mouse Cell Screening (PE) Kit Plate #3	1 plate	96-well	750002446
Cell Staining Buffer	1 bottle	500 mL	420201
Fixation Buffer	1 bottle	100 mL	420801
Plate Sealers	8 sheets	79.4mm x 141mm	78101

Materials to be Provided by the End User

- Adjustable multichannel pipettes for measuring volumes ranging from 25 µL to 1,000 µL
- Centrifuge with a rotor and adaptors for 96-well plates
- Cell culture medium (for cell culture)
- Cell dissociation buffer (for adherent cells)
- 1X PBS (Phosphate-Buffered Saline): 8.0 g NaCl, 1.16 g Na₂HPO₄, 0.2 g KH₂PO₄, 0.2 g KCl, add deionized water to 1 liter; pH to 7.4, 0.2 µm filtered
- Plastic reservoirs for pipetting deionized water or Cell Staining or Fixation Buffer with a multichannel pipette
- A flow cytometer, preferably compatible with reading 96-well plates

Storage and Handling

1. Store unopened kit components at 2 - 8°C. Do not use this kit beyond its expiration date.
2. Once opened, reconstitute plates with distilled, deionized water. The reconstituted plates can be used immediately or sealed for storage at 2 - 8°C in the dark for up to one month.
3. Keep the buffer/s at 2 - 8°C and use within one month after opening.

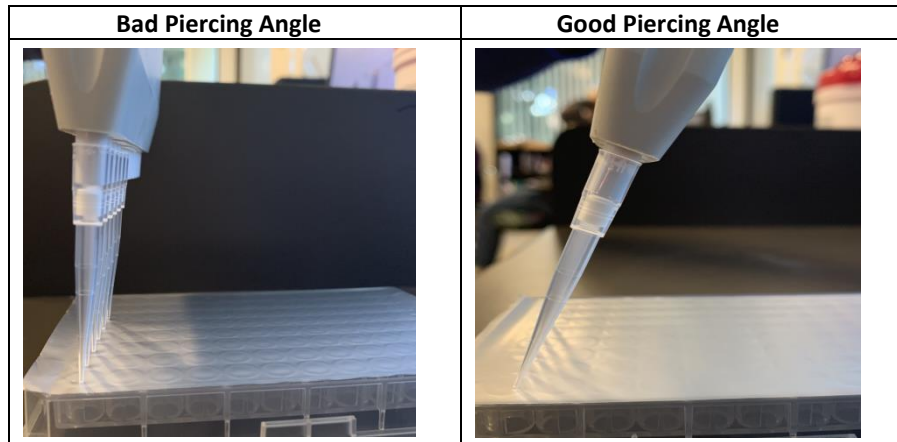
Preparation of Cells for Staining

1. Obtain desired tissue (*e.g. spleen, lymph node, thymus, bone marrow*) and prepare a single cell suspension. Wash cells in 1X PBS or cell culture medium of choice, and resuspend in Cell Staining Buffer at a density between 4×10^6 and 1×10^7 cells/mL.
2. For cultured cells in suspension, spin and resuspend cells in Cell Staining Buffer at a density between 4×10^6 and 1×10^7 cells/mL.
3. For cultured adherent cells, dissociate cells using a mild enzyme or non-enzymatic dissociation buffer. Wash cells in 1X PBS or cell culture medium of choice, and resuspend in Cell Staining Buffer at a density between 4×10^6 and 1×10^7 cells/mL.
4. Filter the cells through a 40 μ m cell strainer to remove any clumps. Keep the cells on ice before use. Approximately 22.5 mL of cells at a density between 4×10^6 and 1×10^7 cells/mL are needed for all three plates. Lower cell density (*e.g. 1.5×10^6 cells/mL*) might be used depending on the application.
5. Optional: Reagents that block Fc receptors (*TruStain fcX™ (anti-mouse CD16/32) Antibody, Cat. No. 101320, BioLegend*) may be useful for reducing nonspecific immunofluorescent staining.

Plate Preparation

One hour before the staining, perform the following steps to prepare the plates.

1. Remove the lyophilized plates from the aluminum pouches. The foil seal can be easily pierced. Handle plates with care by holding along the sides of the plate, once they have been removed from the pouches.
2. Centrifuge the plates at $600 \times g$ for 10 minutes.
3. Ensure the lyophilized cakes have settled to the bottom of the plates. Keep the plates upright at all times from this point forward. Handle plates with care so that the cakes are not agitated at any time.
4. Fill a reservoir (Fisher Scientific Cat# 14-387-069 or equivalent) with deionized water. To reconstitute the lyophilized antibodies, fill the multi-channel pipette with 25 μL /well of deionized water and pierce the plate seal at a 45° angle (see images below for example of good angle and bad angle for piercing the plate seal). This makes it easier to pierce the plate seal. At a 90° angle, more force is needed to pierce the seal.



5. Dispense 25 μL of deionized water **very slowly** to each well, being careful not to cause well contents to splash back onto the film. To avoid cross- contamination, discard the tips after one row or column is reconstituted. Incubate the plate at ambient temperature for 10-15 minutes before removing the seal.
6. Hold the plate firmly then slowly and carefully remove the plate seal, starting from one corner, uncovering one well at a time, for easy opening and to prevent cross-well contamination. You may need to wiggle the seal a little while pulling back slowly.
7. **Discard the plate seal.**
8. Proceed with staining procedure (see next section).
9. If staining cannot be performed immediately, seal the plates with the clear plate sealers provided in the kit.
10. Keep the plates in dark until ready to use.

Note:

- PE-conjugated antibodies are light-sensitive. Try to minimize the exposure of the plates to light as much as practically possible.
- Do not open the pouches until the day you are ready to run the experiment. Once the plates are removed from the pouches, the antibodies must be reconstituted immediately.
- If an experiment is not performed after reconstitution, plates can be sealed and stored in the dark at 2 - 8°C for up to one month.

Cell Staining Procedure

1. Using a multichannel pipette, add 75 μL of cells ($\sim 3 - 7.5 \times 10^5$ cells/well) to each well of the plates.
2. Set up extra tubes to stain cells for flow cytometer setup and compensation, if needed.
3. Using the multichannel pipette, gently mix the cell suspensions by pipetting up and down 2 - 3 times. **Be sure to change tips between each row or column.** Avoid creating bubbles while pipetting.
4. Incubate for 20 - 30 minutes at 2 - 8°C in the dark.
5. Spin the plates at $500 \times g$ for 10 minutes to pellet cells in each well. Immediately after centrifugation, dump the supernatant into the sink by quickly inverting and flicking the plate. Gently blot the plate on a clean paper towel, being careful not to disturb the cell pellet.
6. Using a multi-channel pipette add 200 μL of Cell Staining Buffer to each well. Gently mix up and down to resuspend cells. **Be sure to change tips between each row.**
7. Repeat step 5.
8. To fix the cells, using a multichannel pipette, aliquot 100 μL of Fixation Buffer into each well. Gently mix up and down to resuspend cells. **Be sure to change tips between each row.**
9. Incubate for 10 minutes at room temperature in the dark.
10. Repeat steps 5 - 6.
11. Repeat step 5 one more time.
12. Resuspend cells completely in 160 μL of Cell Staining Buffer per well and analyze using a flow cytometer. We recommend acquiring 70 μL of sample and collecting 5-10, 000 events. Users should determine the optimal number of events to be collected based on specific application they are testing. While the first plate is being acquired, store the other plates at 2 - 8°C in the dark.

Tips for Successful Staining

1. Read the entire manual carefully before the experiment.
2. Plan the experiment in advance. Designate a full day for this experiment. Do not rush any step.
3. Make sure that the flow cytometer's autosampler is well maintained and working well before the experiment. If the flow cytometer does not have an autosampler, the samples from each well of the plates should be transferred to individual FACS tubes and tube adjust the sample volume to approximately 300 μL or more to avoid running dry. Alternatively, a Cluster Tube System (such as Corning catalog # 4410) and acquired manually. The cluster tube can then be transferred to a regular FACS tube for acquisition.
4. Make sure that enough cells have been prepared for the staining. If there are not enough cells, you may choose to divide the staining into two separate experiments.
5. Depending on the application, the cell number needed for staining can be decreased. Successful staining has been done with 1×10^5 cells/well.
6. Make sure to prepare cells for machine setup and compensation. These cells should be treated the same way as the cells for staining in the plates.
7. Handle the plates with care. Keep the plates upright at all times and be careful not to knock the plates over.
8. Protect the plates from exposure to light as much as possible.
9. Use care when handling the plates before reconstitution. The foil seal can be easily pierced. When reconstituting cakes ensure the pipette tips are at a 45° angle to the wells (**see Plate Preparation for instructions**).
10. Use extra caution when removing the plate seal after reconstitution. With the plate on a flat surface, firmly grasp the foil seal at one corner and slowly remove the seal, one well at a time. This will prevent well to well mixing of the reconstituted product (**see Plate Preparation for instructions**).

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11. Some cell surface markers are sensitive to enzymatic digestion. If adherent cells are being used for staining, a mild enzyme or non-enzymatic dissociation buffer should be used when possible.
12. Make sure cells are in a single cell suspension. DNase treatment is recommended to avoid clumps caused by dead cells followed by filtration through a 40 µm cell strainer.
13. Acquire only 70 µL of the 160 µL total volume so that a second run can be performed if necessary.

Data Analysis

We recommend labeling acquisition files as Plate 1_A1, Plate 1_A2 and so on. After sample acquisition is complete, transfer the files into FlowJo™, or equivalent analysis software. Using the “Create Group” function, group samples based on the isotype. This will help set gates for all samples of the same isotype. Antibodies are arranged by isotypes (isotype control followed by all antibodies of that isotype) – for example, Plate 1, A2 is Armenian hamster IgG isotype control and antibodies A3 through E1 are all of the Armenian hamster IgG isotype. Plate 1, E2 is mouse IgG2a isotype control and antibodies E3 through E8 are of the mouse IgG2a isotype control.

Create 11 groups based on the different isotype control antibodies as described below –

1. Armenian Hamster IgG isotype – Plate 1_A2-E1
2. Mouse IgG2a, κ isotype – Plate 1_E2-E8
3. Mouse IgG1, κ isotype – Plate 1_E9-F7
4. Mouse IgG2b, κ isotype – Plate 1_F8-F11
5. Mouse IgM, κ isotype – Plate 1_F12-G1
6. Rat IgG1, κ isotype – Plate 1_G2-H12 and Plate 2_A2-A7
7. Rat IgG2a, κ isotype – Plate 2_A8-H12 and Plate 3_A2-B7
8. Rat IgG2b, κ isotype – Plate 3_B8-F2
9. Rat IgG2c, κ isotype – Plate 3_F3-F5
10. Rat IgM, κ isotype – Plate 3_F6-F8
11. Syrian Hamster IgGκ isotype – Plate 3_F9-G3

Once the groups are created in FlowJo™, go to the first group and gate around the population(s) of interest and then make a histogram plot for PE. Using the isotype control sample, set a gate on the positive population and then apply it to all files in that group. Repeat this process for the remaining 10 groups. Add statistics for percentage of positive, median fluorescence intensity (MFI), event count and any other statistic of interest. Import into an excel file for further analysis using the table editor function.

If comparing control and test samples, then the histograms can be overlaid for visual comparison. If using software other than FlowJo™ please follow a similar strategy.

Frequently Asked Questions

Q: What is the level of variability from one experiment to the other?

A: If the protocol is followed the variability should be minimal. The variability should be similar to single vial antibody staining.

Q: How should the kit be stored?

A: The kit should be stored at 2 - 8°C upon receipt. Once opened, the plates must be reconstituted immediately. Reconstituted plates can be used or stored at 2 - 8°C sealed in the dark and used within a month.

Q: How do I request a custom LEGENDScreen™ product with only my specificities of interest?

A: For more info, visit: biolegend.com/custom_solutions

Q: What are the guarantees regarding the lyophilized plate compared to the reconstituted plate?

A: Lyophilized product has a guaranteed shelf life of 6 months unopened. Reconstituted plates can be used or stored at 2 - 8°C sealed in the dark and used within a month. Be sure to properly seal the plates to prevent evaporation and shield the antibodies from light.

Q: I have added my own antibody solution to the lyophilized product, will the lyophilized antibody work?

A: Yes, as long as the fluorophores on these antibodies are compatible and proper compensation has been applied during acquisition and analysis.

Q: I am not going to use all the reconstituted antibody solution. Can I keep the leftover for later or re-dry the solution in the dark?

A: The antibody is in a one test per well format. There will not be any antibody left if the full test is used. Customers may decide to use less than the recommended volume per test, but this is not recommended and the performance is not guaranteed. Customers may also selectively transfer certain antibodies from the original plate to a new plate and use after reconstitution. If any antibody is not used after reconstitution, the plate can be sealed and store at 2 - 8°C for a month in the dark. Once reconstituted, re-drying is not recommended, as this may result in a loss of signal.

Q: If I don't have enough cells and use less than 4×10^6 /ml (3×10^5 cells/well), will it still work?

A: The kit may work with lower numbers of total cells, but we recommend trying to keep higher concentrations of cells for faster analysis. Of course, how many cells are needed depends on the specific application. Successful staining has been done with 1×10^5 cells/well.

Q: Are these plates made under sterile conditions?

A: The plates are not sterile. Handle them as you would handle a typical flow cytometry staining protocol or reagent.

Q: Can I use half or less of the plate and keep the rest for later?

A: Yes. Customers can use half of the plate or whatever specificities they are interested in. However, the whole plate should be reconstituted. The half plate of antibodies must be transferred to another empty plate for the staining. The remaining half must be sealed and stored at 2 - 8°C in the dark and used within a month.

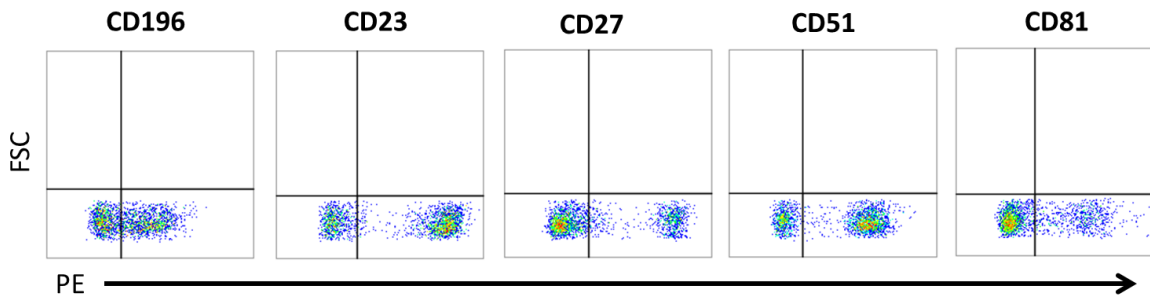
Product Performance

The LEGENDScreen™ Mouse (PE) Kit was tested and compared with BioLegend's cataloged single vial liquid antibody reagents. For cell staining, cells were isolated from spleen and lymph nodes, and 3×10^5 cells were added to each well after the lyophilized antibodies were reconstituted. The cells were then stained for 20 minutes at 4°C, washed, and fixed with Fixation Buffer. The cells were then washed, resuspended in 160 µL of Cell Staining Buffer, and analyzed using an iCyt Eclipse Flow Cytometry Analyzer.

There was no significant difference in the staining patterns or median fluorescence intensity between the lyophilized product and liquid antibodies. Below are some representative data obtained when comparing the LEGENDScreen™ Mouse (PE) Kit vs. equivalent liquid antibodies.

Representative data

Liquid Reagents



LegendScreen Reagents

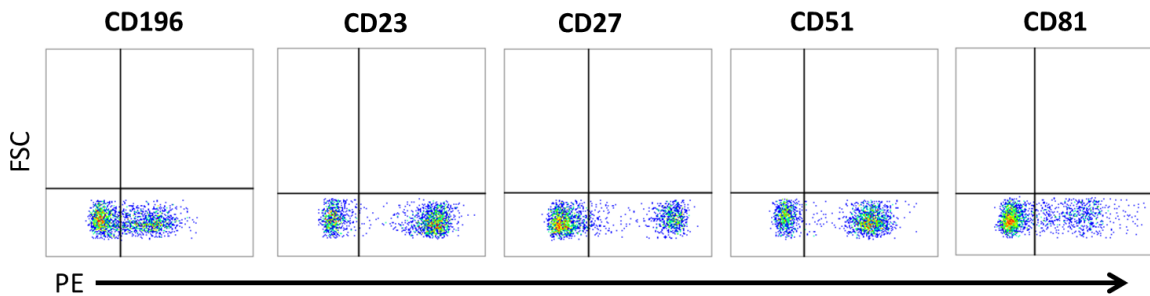


Plate Maps

Plate 1

	1	2	3	4	5	6	7	8	9	10	11	12
A	Blank	Armenian Hamster IgG Isotype Ctrl	CD3ε	CD80	CD81	Notch 1	CD30	CD178 (FasL)	CD103	DLL4	CD195 (CCR5)	Notch 4
B	CD69	Notch 3	JAML	Notch 2	CD194 (CCR4)	CD152	CD120a (TNF R Type I/p55)	CD11c	DLL1	CD196 (CCR6)	CD29	CD55 (DAF)
C	Jagged 2	CD79b (Igβ)	IFN-γR β chain	CD61	CD121a (IL-1 R, Type I/p80)	TCR β chain	FcεR1α	CD16.2 (FcγRIV)	CD36	DcTRAIL-R1	CD84	CD48
D	CD49b	CD120b (TNF R Type II/p75)	CD262 (DR5, TRAIL-R2)	CD270 (HVEM)	TCR Vγ1.1 + Vγ1.2	B7-H4 (B7S1, B7X)	CD339 (Jagged 1)	CD49a	CD85k (gp49 Receptor)	Plexin B2	CD27	DR3 (TNFRSF25)
E	TCR γ/δ	Mouse IgG2a, κ Isotype Ctrl	CD45.1	CD45.2	NK-1.1	Ly108	CD207 (Langerin)	CX3CR1	Mouse IgG1, κ Isotype Ctrl	CD66a (CEACAM1a)	IFNAR-1	Tim-2
F	CD272 (BTLA)	CD64 (FcγRI)	CD351 (Fc α/μ receptor)	LAP (TGF-β1)	CD59a	Ly49H	CD90/CD90.1 (Thy-1.1)	Mouse IgG2b, κ Isotype Ctrl	CD157 (BST-1)	CD159a (NKG2AB6)	XCR1	Mouse IgM, κ Isotype Ctrl
G	CD15 (SSEA-1)	Rat IgG1, κ Isotype Ctrl	Trem-like 4 (Trem14)	Ig light chain κ	Siglec H	CD255 (TWEAK)	CD202b (Tie-2, CD202)	GITR Ligand	CD147	CD73	CD51	CD314 (NKG2D)
H	CD96 (TACTILE)	FR4 (Folate Receptor 4)	CD210 (IL-10 R)	CD83	CD107b (Mac-3)	CD223 (LAG-3)	CD134 (OX-40)	CD117 (c-kit)	CD41	CD268 (BAFF-R)	CD144 (VE-cadherin)	CD370 (CLEC9A, DNGR1)

Plate 2

	1	2	3	4	5	6	7	8	9	10	11	12
A	Blank	CD369 (Dectin-1 /CLEC7A)	PIR-A/B	CD22	CD324 (E-Cadherin)	CD172a (SIRPα)	CD319	Rat IgG2a, κ Isotype Ctrl	MAIR-V	CD146	PD-1H (VISTA)	CD8a
B	CD275 (B7-H2, B7- RP1, ICOS Ligand)	Ly-6A/E (Sca-1)	CD40	CD45R/ B220	CD197 (CCR7)	CD47	CD98 (4F2)	CD14	CD107a (LAMP-1)	CD18	Ly-6G	CD21/CD35 (CR2/CR1)
C	Mac-2 (Galectin-3)	CD199 (CCR9)	Ly-51	IgD	Tim-4	CD71	H-2	CD45RB	CD326 (Ep-CAM)	IgM	CD155 (PVR)	CD200R (OX2R)
D	CD254 (TRANCE, RANKL)	IL-21R	CD276 (B7-H3)	CD9	CD105	CD366 (Tim-3)	4-1BB Ligand (CD137L)	CD265 (RANK)	TLR4 (CD284)/ MD2 Complex	CD19	LPAM-1 (Integrin α4β7)	CD62L
E	CD23	CD5	CD273 (B7-DC, PD-L2)	F4/80	CD94	CD267 (TAC1)	Ly-49A	CD180 (RP105)	CD11a	lymphotoxin beta receptor (LTβR)	CD122 (IL-2Rβ)	CD106
F	integrin β7	CD115 (CSF-1R)	CD140a	PDC-TREM	CD135	CD140b	ESAM	CD200 (OX2)	CD309 (VEGFR2, Flk-1)	TLT-2	CD253 (TRAIL)	CD335 (NKp46)
G	CD205 (DEC-205)	Galectin-9	CD200R3	MAIR-IV (CLM-5)	Ly49D	CD123	CD355 (CRTAM)	CD169 (Siglec-1)	CD138 (Syndecan-1)	CD160	CD39	GARP (LRRC32)
H	CD179a (VpreB)	CD371 (CLEC12A)	CD63	CD49e	CD193 (CCR3)	CD300LG (Nepmucin)	CD301a (MGL1)	IL-33Rα (IL1RL1, ST2)	CD304 (Neuropilin-1)	CD6	CD100 (SEMA4D)	CD104

Plate 3

	1	2	3	4	5	6	7	8	9	10	11	12
A	Blank	CD182 (CXCR2)	MAdCAM-1	MERTK (Mer)	CD226 (DNAM-1)	Ly6K	CD150 (SLAM)	CD25	CD38	CD133	CD301b (MGL2)	CD34
B	CD154 (CD40L)	TIGIT (Vstm3)	CD126 (IL-6R α)	CD86	CD37	CD365 (Tim-1)	PD-1H (VISTA)	Rat IgG2b, κ Isotype Ctrl	CD70	CD4	I-A/I-E	CD153
C	CD54	DC Marker (33D1)	CD90.2	TER-119/ Erythroid Cells	CD49d	CD24	Ly-6G/ Ly-6C (Gr-1)	CD1d (CD1.1, Ly-38)	CD11b	CD45	RAE-1 γ	CD8b
D	CD44	CD43	CD317 (BST2, PDCA-1)	CD132 (common γ chain)	CD3	CD274 (B7-H1, PD-L1)	CD88 (C5aR)	CD93 (AA4.1, early B lineage)	CD252 (OX40L)	MD-1	CD357 (GITR)	CD185 (CXCR5)
E	CD300c/d	CD186 (CXCR6)	CD130 (gp130)	CD198 (CCR8)	CD20	CD124 (IL-4R α)	IL-23R	CD184 (CXCR4)	CD2	CD183 (CXCR3)	CD16/32	CD279 (PD-1)
F	CD31 (PECAM-1)	CD127 (IL-7R α)	Rat IgG2c, κ Isotype Ctrl	Ly-6C	Ly-6D	Rat IgM, κ Isotype Ctrl	CD49b (pan-NK cells)	GL7 Antigen (T and B cell Activation Marker)	Syrian Hamster IgG Isotype Ctrl	CD28	Podoplanin	CD137
G	CD278 (ICOS)	KLRG1 (MAFA)	Ly-49C/ F/I/H	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank
H	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank	Blank

Antibody Information Table

Plate 1

Well	Cat #	Clone	Description	Isotype
A1			Blank	
A2	400908	HTK888	Armenian Hamster IgG Isotype Ctrl	Armenian Hamster IgG
A3	100308	145-2C11	anti-mouse CD3ε	Armenian Hamster IgG
A4	104708	16-10A1	anti-mouse CD80	Armenian Hamster IgG
A5	104906	Eat-2	anti-mouse/rat CD81	Armenian Hamster IgG
A6	130608	HMN1-12	anti-mouse Notch 1	Armenian Hamster IgG
A7	102306	mCD30.1	anti-mouse CD30	Armenian Hamster IgG
A8	106606	MFL3	anti-mouse CD178 (FasL)	Armenian Hamster IgG
A9	121405	2E7	anti-mouse CD103	Armenian Hamster IgG
A10	130808	HMD4-1	anti-mouse DLL4	Armenian Hamster IgG
A11	107006	HM-CCR5	anti-mouse CD195 (CCR5)	Armenian Hamster IgG
A12	128407	HMN4-14	anti-mouse Notch 4	Armenian Hamster IgG
B1	104508	H1.2F3	anti-mouse CD69	Armenian Hamster IgG
B2	130507	HMN3-133	anti-mouse Notch 3	Armenian Hamster IgG
B3	128503	4E10	anti-mouse JAML	Armenian Hamster IgG
B4	130707	HMN2-35	anti-mouse Notch 2	Armenian Hamster IgG
B5	131204	2G12	anti-mouse CD194 (CCR4)	Armenian Hamster IgG

Well	Cat #	Clone	Description	Isotype
E1	118108	GL3	anti-mouse TCR γ/δ	Armenian Hamster IgG
E2	400212	MOPC-173	Mouse IgG2a, κ Isotype Ctrl	Mouse IgG2a, κ
E3	110708	A20	anti-mouse CD45.1	Mouse IgG2a, κ
E4	109808	104	anti-mouse CD45.2	Mouse IgG2a, κ
E5	156503	S17016D	anti-mouse NK-1.1	Mouse IgG2a, κ
E6	134606	330-AJ	anti-mouse Ly108	Mouse IgG2a, κ
E7	144204	4C7	anti-mouse/human CD207 (Langerin)	Mouse IgG2a, κ
E8	149006	SA011F11	anti-mouse CX3CR1	Mouse IgG2a, κ
E9	400112	MOPC-21	Mouse IgG1, κ Isotype Ctrl	Mouse IgG1, κ
E10	134506	MAb-CC1	anti-mouse CD66a (CEACAM1a)	Mouse IgG1, κ
E11	127312	MAR1-5A3	anti-mouse IFNAR-1	Mouse IgG1, κ
E12	129006	F37-2C4	anti-mouse Tim-2	Mouse IgG1, κ
F1	134804	8F4	anti-mouse CD272 (BTLA)	Mouse IgG1, κ
F2	139304	X54-5/7.1	anti-mouse CD64 (FcγRI)	Mouse IgG1, κ
F3	137306	TX61	anti-mouse/human CD351 (Fc α/μ receptor)	Mouse IgG1, κ
F4	141306	TW7-20B9	anti-mouse LAP (TGF-β1)	Mouse IgG1, κ
F5	143104	mCD59.3	anti-mouse CD59a	Mouse IgG1, κ

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Well	Cat #	Clone	Description	Isotype
B6	106306	UC10-4B9	anti-mouse CD152	Armenian Hamster IgG
B7	113004	55R-286	anti-mouse CD120a (TNF R Type I/p55)	Armenian Hamster IgG
B8	117308	N418	anti-mouse CD11c	Armenian Hamster IgG
B9	128307	HMD1-3	anti-mouse DLL1	Armenian Hamster IgG
B10	129804	29-2L17	anti-mouse CD196 (CCR6)	Armenian Hamster IgG
B11	102208	HMβ1-1	anti-mouse/rat CD29	Armenian Hamster IgG
B12	131804	RIKO-3	anti-mouse CD55 (DAF)	Armenian Hamster IgG
C1	131007	HMJ2-1	anti-mouse Jagged 2	Armenian Hamster IgG
C2	132804	HM79-12	anti-mouse CD79b (Igb)	Armenian Hamster IgG
C3	113604	MOB-47	anti-mouse IFN-γR β chain	Armenian Hamster IgG
C4	104308	2C9.G2 (HMβ3-1)	anti-mouse/rat CD61	Armenian Hamster IgG
C5	113505	JAMA-147	anti-mouse CD121a (IL-1 R, Type I/p80)	Armenian Hamster IgG
C6	109208	H57-597	anti-mouse TCR β chain	Armenian Hamster IgG
C7	134308	MAR-1	anti-mouse FcεR1α	Armenian Hamster IgG
C8	149503	9E9	anti-mouse CD16.2 (FcγRIV)	Armenian Hamster IgG
C9	102606	HM36	anti-mouse CD36	Armenian Hamster IgG
C10	133804	mDcR1-3	anti-mouse DcTRAIL-R1	Armenian Hamster IgG
C11	122806	mCD84.7	anti-mouse CD84	Armenian Hamster IgG
C12	103406	HM48-1	anti-mouse CD48	Armenian Hamster IgG

Well	Cat #	Clone	Description	Isotype
F6	144706	3D10	anti-mouse Ly49H	Mouse IgG1, κ
F7	202523	OX-7	anti-rat CD90/mouse CD90.1 (Thy-1.1)	Mouse IgG1, κ
F8	400312	MPC-11	Mouse IgG2b, κ Isotype Ctrl	Mouse IgG2b, κ
F9	140204	BP-3	anti-mouse CD157 (BST-1)	Mouse IgG2b, κ
F10	142804	16A11	anti-mouse CD159a (NKG2AB6)	Mouse IgG2b, κ
F11	148204	ZET	anti-mouse/rat XCR1	Mouse IgG2b, κ
F12	401611	MM-30	Mouse IgM, κ Isotype Ctrl	Mouse IgM, κ
G1	125606	MC-480	anti-mouse/human CD15 (SSEA-1)	Mouse IgM, κ
G2	400408	RTK2071	Rat IgG1, κ Isotype Ctrl	Rat IgG1, κ
G3	143303	16E5	anti-mouse Trem-like 4 (Trem14)	Rat IgG1, κ
G4	409506	RMK-45	anti-mouse Ig light chain κ	Rat IgG
G5	129606	551	anti-mouse Siglec H	Rat IgG1, κ
G6	120005	MTW-1	anti-mouse CD255 (TWEAK)	Rat IgG1, κ
G7	124008	TEK4	anti-mouse CD202b (Tie-2, CD202)	Rat IgG1, κ
G8	120306	YGL 386	anti-mouse GITR Ligand	Rat IgG1, κ
G9	123707	OX-114	anti-mouse CD147	Rat IgG1, κ
G10	127206	TY/11.8	anti-mouse CD73	Rat IgG1, κ
G11	104106	RMV-7	anti-mouse CD51	Rat IgG1, κ
G12	130208	CX5	anti-mouse CD314 (NKG2D)	Rat IgG1, κ

LEGENDScreen™ Mouse (PE) Kit

Well	Cat #	Clone	Description	Isotype
D1	103506	HMa2	anti-mouse CD49b	Armenian Hamster IgG
D2	113406	TR75-89	anti-mouse CD120b (TNF R Type II/p75)	Armenian Hamster IgG
D3	119906	MD5-1	anti-mouse CD262 (DR5, TRAIL-R2)	Armenian Hamster IgG
D4	136304	HMHV-1B18	anti-mouse CD270 (HVEM)	Armenian Hamster IgG
D5	142704	4B2.9	anti-mouse TCR Vy1.1 + Vy1.2	Armenian Hamster IgG
D6	139406	HMH4-5G1	anti-mouse B7-H4 (B7S1, B7X)	Armenian Hamster IgG
D7	130908	HMJ1-29	anti-mouse CD339 (Jagged 1)	Armenian Hamster IgG
D8	142604	HMa1	anti-mouse CD49a	Armenian Hamster IgG
D9	144904	H1.1	anti-mouse CD85k (gp49 Receptor)	Armenian Hamster IgG
D10	145903	3E7	anti-mouse Plexin B2	Armenian Hamster IgG
D11	124210	LG.3A10	anti-mouse/rat/human CD27	Armenian Hamster IgG
D12	144406	4C12	anti-mouse DR3 (TNFRSF25)	Armenian Hamster IgG

Well	Cat #	Clone	Description	Isotype
H1	131705	3.3	anti-mouse CD96 (TACTILE)	Rat IgG1, κ
H2	125007	12A5	anti-mouse FR4 (Folate Receptor 4)	Rat IgG1, κ
H3	112706	1B1.3a	anti-mouse CD210 (IL-10 R)	Rat IgG1, κ
H4	121508	Michel-19	anti-mouse CD83	Rat IgG1, κ
H5	108506	M3/84	anti-mouse CD107b (Mac-3)	Rat IgG1, κ
H6	125208	C9B7W	anti-mouse CD223 (LAG-3)	Rat IgG1, κ
H7	119410	OX-86	anti-mouse CD134 (OX-40)	Rat IgG1, κ
H8	161503	S18020A	anti-mouse CD117 (c-kit)	Rat IgG1, κ
H9	133906	MWReg30	anti-mouse CD41	Rat IgG1, κ
H10	134104	7H22-E16	anti-mouse CD268 (BAFF-R)	Rat IgG1, κ
H11	138010	BV13	anti-mouse CD144 (VE-cadherin)	Rat IgG1, κ
H12	143504	7H11	anti-mouse CD370 (CLEC9A, DNCR1)	Rat IgG1, κ

Plate 2

Well	Cat #	Clone	Description	Isotype
A1			Blank	
A2	144304	RH1	anti-mouse CD369 (Dectin-1/CLEC7A)	Rat IgG1, κ
A3	144104	6C1	anti-mouse PIR-A/B	Rat IgG1, κ
A4	126112	OX-97	anti-mouse CD22	Rat IgG1, κ
A5	147304	DECMA-1	anti-mouse/human CD324 (E-Cadherin)	Rat IgG1, κ
A6	144012	P84	anti-mouse CD172a (SIRPα)	Rat IgG1, κ
A7	152006	4G2	anti-mouse CD319	Rat IgG1, κ
A8	400508	RTK2758	Rat IgG2a, κ Isotype Ctrl	Rat IgG2a, κ
A9	132704	TX70	anti-mouse MAIR-V	Rat IgG2a, κ
A10	134704	ME-9F1	anti-mouse CD146	Rat IgG2a, κ
A11	159603	A172071	anti-mouse PD-1H (VISTA)	Rat IgG2a, κ
A12	100708	53-6.7	anti-mouse CD8a	Rat IgG2a, κ
B1	107405	HK5.3	anti-mouse CD275 (B7-H2, B7-RP1, ICOS Ligand)	Rat IgG2a, κ
B2	122507	E13-161.7	anti-mouse Ly-6A/E (Sca-1)	Rat IgG2a, κ
B3	124610	3/23	anti-mouse CD40	Rat IgG2a, κ
B4	103208	RA3-6B2	anti-mouse/human CD45R/B220	Rat IgG2a, κ
B5	120106	4B12	anti-mouse CD197 (CCR7)	Rat IgG2a, κ

Well	Cat #	Clone	Description	Isotype
E1	101608	B3B4	anti-mouse CD23	Rat IgG2a, κ
E2	100608	53-7.3	anti-mouse CD5	Rat IgG2a, κ
E3	107206	TY25	anti-mouse CD273 (B7-DC, PD-L2)	Rat IgG2a, κ
E4	123110	BM8	anti-mouse F4/80	Rat IgG2a, κ
E5	105508	18d3	anti-mouse CD94	Rat IgG2a, κ
E6	133404	8F10	anti-mouse CD267 (TACI)	Rat IgG2a, κ
E7	116808	YE1/48.10.6	anti-mouse Ly-49A	Rat IgG2a, κ
E8	117706	RP/14	anti-mouse CD180 (RP105)	Rat IgG2a, κ
E9	101107	M17/4	anti-mouse CD11a	Rat IgG2a, κ
E10	134403	5G11	anti-mouse lymphotoxin beta receptor (LTβR)	Rat IgG2a, κ
E11	105906	5H4	anti-mouse CD122 (IL-2RB)	Rat IgG2a, κ
E12	105713	429 (MVCAM.A)	anti-mouse CD106	Rat IgG2a, κ
F1	121006	FIB27	anti-human/mouse integrin β7	Rat IgG2a, κ
F2	135506	AFS98	anti-mouse CD115 (CSF-1R)	Rat IgG2a, κ
F3	135906	APA5	anti-mouse CD140a	Rat IgG2a, κ
F4	139204	4A6	anti-mouse PDC-TREM	Rat IgG2a, κ
F5	135306	A2F10	anti-mouse CD135	Rat IgG2a, κ

LEGENDScreen™ Mouse (PE) Kit

Well	Cat #	Clone	Description	Isotype
B6	127508	miap301	anti-mouse CD47	Rat IgG2a, κ
B7	128208	RL388	anti-mouse CD98 (4F2)	Rat IgG2a, κ
B8	123310	Sa14-2	anti-mouse CD14	Rat IgG2a, κ
B9	121612	1D4B	anti-mouse CD107a (LAMP-1)	Rat IgG2a, κ
B10	101408	M18/2	anti-mouse CD18	Rat IgG2a, κ
B11	164503	S19018G	anti-mouse Ly-6G	Rat IgG2a, κ
B12	123410	7E9	anti-mouse CD21/CD35 (CR2/CR1)	Rat IgG2a, κ
C1	125405	M3/38	anti-mouse/human Mac-2 (Galectin-3)	Rat IgG2a, κ
C2	129708	9B1	anti-mouse CD199 (CCR9)	Rat IgG2a, κ
C3	108308	6C3	anti-mouse Ly-51	Rat IgG2a, κ
C4	405706	11-26c.2a	anti-mouse IgD	Rat IgG2a, κ
C5	130006	RMT4-54	anti-mouse Tim-4	Rat IgG2a, κ
C6	113808	RI7217	anti-mouse CD71	Rat IgG2a, κ
C7	125506	M1/42	anti-mouse H-2	Rat IgG2a, κ
C8	103308	C363-16A	anti-mouse CD45RB	Rat IgG2a, κ
C9	118206	G8.8	anti-mouse CD326 (Ep-CAM)	Rat IgG2a, κ
C10	406508	RMM-1	anti-mouse IgM	Rat IgG2a, κ
C11	131508	TX56	anti-mouse CD155 (PVR)	Rat IgG2a, κ
C12	123908	OX-110	anti-mouse CD200R (OX2R)	Rat IgG2a, κ

Well	Cat #	Clone	Description	Isotype
F6	136006	APB5	anti-mouse CD140b	Rat IgG2a, κ
F7	136203	1G8/ESAM	anti-mouse ESAM	Rat IgG2a, κ
F8	123808	OX-90	anti-mouse CD200 (OX2)	Rat IgG2a, κ
F9	136404	Avas12	anti-mouse CD309 (VEGFR2, Flk-1)	Rat IgG2a, κ
F10	136603	MIH47	anti-mouse TLT-2	Rat IgG2a, κ
F11	109306	N2B2	anti-mouse CD253 (TRAIL)	Rat IgG2a, κ
F12	137604	29A1.4	anti-mouse CD335 (NKP46)	Rat IgG2a, κ
G1	138213	NLDC-145	anti-mouse CD205 (DEC-205)	Rat IgG2a, κ
G2	137904	108A2	anti-mouse Galectin-9	Rat IgG2a, κ
G3	142206	Ba13	anti-mouse CD200R3	Rat IgG2a, κ
G4	139605	TX69	anti-mouse MAIR-IV (CLM-5)	Rat IgG2a, κ
G5	138308	4E5	anti-mouse Ly49D	Rat IgG2a, κ
G6	106005	5B11	anti-mouse CD123	Rat IgG2a, κ
G7	142005	11-5/CRTAM	anti-mouse CD355 (CRTAM)	Rat IgG2a, κ
G8	142404	3D6.112	anti-mouse CD169 (Siglec-1)	Rat IgG2a, κ
G9	142504	281-2	anti-mouse CD138 (Syndecan-1)	Rat IgG2a, κ
G10	143004	7H1	anti-mouse CD160	Rat IgG2a, κ
G11	143804	Duha59	anti-mouse CD39	Rat IgG2a, κ
G12	142904	F011-5	anti-mouse GARP (LRRC32)	Rat IgG2a, κ

LEGENDScreen™ Mouse (PE) Kit

Well	Cat #	Clone	Description	Isotype
D1	510006	IK22/5	anti-mouse CD254 (TRANCE, RANKL)	Rat IgG2a, κ
D2	131906	4A9	anti-mouse IL-21R	Rat IgG2a, κ
D3	124508	RTAA15	anti-mouse CD276 (B7-H3)	Rat IgG2a, κ
D4	124806	MZ3	anti-mouse CD9	Rat IgG2a, κ
D5	120408	MJ7/18	anti-mouse CD105	Rat IgG2a, κ
D6	119704	RMT3-23	anti-mouse CD366 (Tim-3)	Rat IgG2a, κ
D7	107105	TKS-1	anti-mouse 4-1BB Ligand (CD137L)	Rat IgG2a, κ
D8	119806	R12-31	anti-mouse CD265 (RANK)	Rat IgG2a, κ
D9	117606	MTS510	anti-mouse TLR4 (CD284)/ MD2 Complex	Rat IgG2a, κ
D10	115508	6D5	anti-mouse CD19	Rat IgG2a, κ
D11	120606	DATK32	anti-mouse LPAM-1 (Integrin α4β7)	Rat IgG2a, κ
D12	161203	W18021D	anti-mouse CD62L	Rat IgG2a, κ

Well	Cat #	Clone	Description	Isotype
H1	143604	R3	anti-mouse CD179a (VpreB)	Rat IgG2a, κ
H2	143403	5D3/CLEC12A	anti-mouse CD371 (CLEC12A)	Rat IgG2a, κ
H3	143904	NVG-2	anti-mouse CD63	Rat IgG2a, κ
H4	103805	5H10-27(MFR5)	anti-mouse CD49e	Rat IgG2a, κ
H5	144506	J073E5	anti-mouse CD193 (CCR3)	Rat IgG2a, κ
H6	147104	ZAQ5	anti-mouse CD300LG (Nepmucin)	Rat IgG2a, κ
H7	145606	LOM-8.7	anti-mouse CD301a (MGL1)	Rat IgG2a, κ
H8	145304	DIH9	anti-mouse IL-33Rα (IL1RL1, ST2)	Rat IgG2a, κ
H9	145204	3E12	anti-mouse CD304 (Neuropilin-1)	Rat IgG2a, κ
H10	146404	OX-129	anti-mouse CD6	Rat IgG2a, κ
H11	147603	BMA-12	anti-mouse CD100 (SEMA4D)	Rat IgG2a, κ
H12	123610	346-11A	anti-mouse CD104	Rat IgG2a, κ

Plate 3

Well	Cat #	Clone	Description	Isotype
A1			Blank	
A2	149304	SA044G4	anti-mouse CD182 (CXCR2)	Rat IgG2a, κ
A3	120710	MECA-367	anti-mouse MAdCAM-1	Rat IgG2a, κ
A4	151506	2B10C42	anti-mouse MERTK (Mer)	Rat IgG2a, κ
A5	133603	TX42.1	anti-mouse CD226 (DNAM-1)	Rat IgG2a, κ
A6	151304	MK34	anti-mouse Ly6K	Rat IgG2a, κ
A7	115903	TC15-12F12.2	anti-mouse CD150 (SLAMF6)	Rat IgG2a, κ
A8	162103	QA19A49	anti-mouse CD25	Rat IgG2a, κ
A9	102708	90	anti-mouse CD38	Rat IgG2a, κ
A10	141204	315-2C11	anti-mouse CD133	Rat IgG2a, λ
A11	146804	URA-1	anti-mouse CD301b (MGL2)	Rat IgG2a, λ
A12	152204	SA376A4	anti-mouse CD34	Rat IgG2a, λ
B1	157003	SA047C3	anti-mouse CD154 (CD40L)	Rat IgG2a, κ
B2	622205	A17200C	anti-mouse TIGIT (Vstm3)	Rat IgG2a, κ
B3	160405	W18166A	anti-mouse CD126 (IL-6Rα)	Rat IgG2a, κ
B4	159203	A17199A	anti-mouse CD86	Rat IgG2a, κ
B5	146204	Duno85	anti-mouse CD37	Rat IgG2a, κ

Well	Cat #	Clone	Description	Isotype
E1	148004	TX52	anti-mouse CD300c/d	Rat IgG2b, κ
E2	151104	SA051D1	anti-mouse CD186 (CXCR6)	Rat IgG2b, κ
E3	149404	4H1B35	anti-mouse CD130 (gp130)	Rat IgG2b, κ
E4	150312	SA214G2	anti-mouse CD198 (CCR8)	Rat IgG2b, κ
E5	150410	SA275A11	anti-mouse CD20	Rat IgG2b, κ
E6	144804	I015F8	anti-mouse CD124 (IL-4Rα)	Rat IgG2b, κ
E7	150904	12B2B64	anti-mouse IL-23R	Rat IgG2b, κ
E8	146506	L276F12	anti-mouse CD184 (CXCR4)	Rat IgG2b, κ
E9	100108	RM2-5	anti-mouse CD2	Rat IgG2b, λ
E10	155903	S18001A	anti-mouse CD183 (CXCR3)	Rat IgG2b, κ
E11	156605	S17011E	anti-mouse CD16/32	Rat IgG2b, κ
E12	109104	RMP1-30	anti-mouse CD279 (PD-1)	Rat IgG2b, κ
F1	160203	W18222B	anti-mouse CD31 (PECAM-1)	Rat IgG2b, κ
F2	158203	S18006K	anti-mouse CD127 (IL-7Rα)	Rat IgG2b, λ
F3	400707	RTK4174	Rat IgG2c, κ Isotype Ctrl	Rat IgG2c, κ
F4	128008	HK1.4	anti-mouse Ly-6C	Rat IgG2c, κ
F5	138604	49-H4	anti-mouse Ly-6D	Rat IgG2c, κ

LEGENDScreen™ Mouse (PE) Kit

Well	Cat #	Clone	Description	Isotype
B6	119506	RMT1-4	anti-mouse CD365 (Tim-1)	Rat IgG2a, κ
B7	159503	A17207C	anti-mouse PD-1H (VISTA)	Rat IgG2a, κ
B8	400608	RTK4530	Rat IgG2b, κ Isotype Ctrl	Rat IgG2b, κ
B9	104606	FR70	anti-mouse CD70	Rat IgG2b, κ
B10	100408	GK1.5	anti-mouse CD4	Rat IgG2b, κ
B11	107607	M5/114.15.2	anti-mouse I-A/I-E	Rat IgG2b, κ
B12	106405	RM153	anti-mouse CD153	Rat IgG2b, κ
C1	116108	YN1/1.7.4	anti-mouse CD54	Rat IgG2b, κ
C2	124905	33D1	anti-mouse DC Marker (33D1)	Rat IgG2b, κ
C3	105308	30-H12	anti-mouse CD90.2	Rat IgG2b, κ
C4	116208	TER-119	anti-mouse TER-119/ Erythroid Cells	Rat IgG2b, κ
C5	103608	R1-2	anti-mouse CD49d	Rat IgG2b, κ
C6	101808	M1/69	anti-mouse CD24	Rat IgG2b, κ
C7	108408	RB6-8C5	anti-mouse Ly-6G/Ly-6C (Gr-1)	Rat IgG2b, κ
C8	123510	1B1	anti-mouse CD1d (CD1.1, Ly-38)	Rat IgG2b, κ
C9	101208	M1/70	anti-mouse/human CD11b	Rat IgG2b, κ
C10	103106	30-F11	anti-mouse CD45	Rat IgG2b, κ
C11	130107	CX1	anti-mouse RAE-1γ	Rat IgG2b, κ
C12	126608	YTS156.7.7	anti-mouse CD8b	Rat IgG2b, κ

Well	Cat #	Clone	Description	Isotype
F6	400808	RTK2118	Rat IgM, κ Isotype Ctrl	Rat IgM, κ
F7	108908	DX5	anti-mouse CD49b (pan-NK cells)	Rat IgM, κ
F8	144608	GL7	anti-mouse/human GL7 Antigen (T and B cell Activation Marker)	Rat IgM, κ
F9	402008	SHG-1	Syrian Hamster IgG Isotype Ctrl	Syrian Hamster IgG
F10	102106	37.51	anti-mouse CD28	Syrian Hamster IgG
F11	127408	8.1.1	anti-mouse Podoplanin	Syrian Hamster IgG
F12	106106	17B5	anti-mouse CD137	Syrian Hamster IgG
G1	107706	15F9	anti-mouse CD278 (ICOS)	Syrian Hamster IgG
G2	138408	2F1/KLRG1	anti-mouse/human KLRG1 (MAFA)	Syrian Hamster IgG
G3	108208	14B11	anti-mouse Ly-49C/F/I/H	Syrian Hamster IgG
G4			Blank	
G5			Blank	
G6			Blank	
G7			Blank	
G8			Blank	
G9			Blank	
G10			Blank	
G11			Blank	
G12			Blank	

LEGENDScreen™ Mouse (PE) Kit

Well	Cat #	Clone	Description	Isotype
D1	103008	IM7	anti-mouse/human CD44	Rat IgG2b, κ
D2	143206	S11	anti-mouse CD43	Rat IgG2b, κ
D3	127010	927	anti-mouse CD317 (BST2, PDCA-1)	Rat IgG2b, κ
D4	132306	TUGm2	anti-mouse CD132 (common γ chain)	Rat IgG2b, κ
D5	100205	17A2	anti-mouse CD3	Rat IgG2b, κ
D6	124307	10F.9G2	anti-mouse CD274 (B7-H1, PD-L1)	Rat IgG2b, κ
D7	135806	20/70	anti-mouse CD88 (C5aR)	Rat IgG2b, κ
D8	136504	AA4.1	anti-mouse CD93 (AA4.1, early B lineage)	Rat IgG2b, κ
D9	108806	RM134L	anti-mouse CD252 (OX40L)	Rat IgG2b, κ
D10	117805	MD-113	anti-mouse MD-1	Rat IgG2b, κ
D11	120208	YGITR 765	anti-mouse CD357 (GITR)	Rat IgG2b, κ
D12	145504	L138D7	anti-mouse CD185 (CXCR5)	Rat IgG2b, κ

Well	Cat #	Clone	Description	Isotype
H1			Blank	
H2			Blank	
H3			Blank	
H4			Blank	
H5			Blank	
H6			Blank	
H7			Blank	
H8			Blank	
H9			Blank	
H10			Blank	
H11			Blank	
H12			Blank	



LEGENDScreen™ kits are manufactured by **BioLegend**

8999 BioLegend Way

San Diego, CA 92121

United States

For contact information, as well as a list of world-wide BioLegend offices and distributors, please visit: biolegend.com/en-us/contact