Product Datasheet

Anti-F4/80 [Cl:A3-1 (recombinant version)] (orb411543)

Description	Rat monoclonal antibody to F4/80
Species/Host	Rat
Reactivity	Mouse
Conjugation	Unconjugated
Tested Applications	FC, IHC
Immunogen	Thioglycollate stimulated peritoneal macrophages of mouse origin.
Target	F4/80
Preservatives	PBS with 0.02% Proclin 300.
Concentration	1 mg/ml
Storage	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.
Note	For research use only
Isotype	IgG2b
Clonality	Monoclonal
Clone Number	Cl:A3-1 (recombinant version)
Antibody Type	Recombinant Antibody
Purity	Purified
Uniprot ID	Q61549

Biorbyt Ltd.
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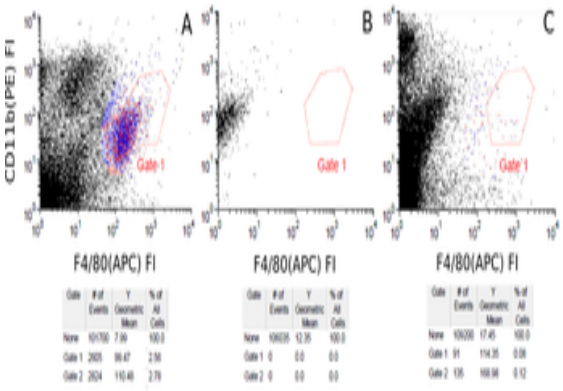
Expiration Date 12 months from date of receipt.



Competitive flow-cytometry assay between anti-F4/80 CI:A3-1 variants and an existing commercial anti-F4/80 antibody. Mouse (Mus musculus) splenocytes were labelled ex vivo with a commercially available APC-labelled anti-F4/80 antibody and APE labelled anti-CD11b antibody and subject to flow-cytometry analysis (A), in which a small subpopulation of F4/80-CD11b positive cells may be observed. Subsets of commerical anti-F4/80 antibody-labelled splenocytes were also subsequently incubated with unlabelled versions of either the rat (Rattus norvegicus) IgG2b chimeric version (orb411543, B) or mouse IgG2A chimeric (C) version of CI:A3-1. Loss of the F4/80-CD11b positive subpopulation may be observed, demonstrating displacement of the commercial antibody and the specificity of CI:A3-1.

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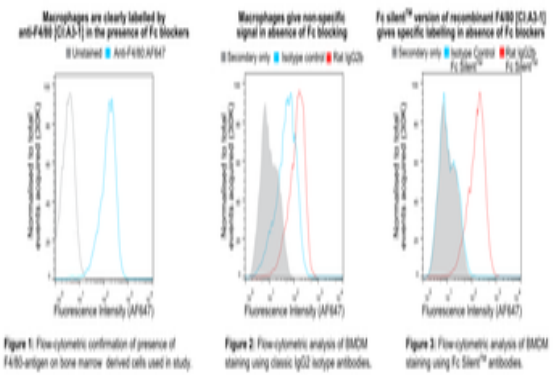

In Figure 1 murine bone marrow-derived macrophages (BMDMs) were pre-blocked with rat anti-mouse CD16 & CD32 (clone FCR-4G8) and stained with non-recombinant anti-F4/80 [Cl:A3-1] conjugated to Alexa Fluor® 647 (AF647), all commercially available from competitors. In Figure 2 BMDMs were stained with recombinant anti-F4/80 [Cl:A3-1] or isotype control (anti-fluorescein [4-4-20 (enhanced)] IgG2b (orb256396). In Figure 3 BMDMs were stained with Fc Silent™ recombinant anti-F4/80 [Cl:A3-1] or isotype control (Fc Silent™ anti-fluorescein [4-4-20 (enhanced)] IgG2b. These were fluorescently labelled using the secondary antibody, goat IgG anti-rat IgG (H&L-chain) polyclonal antibody directly conjugated to Alexa Fluor® 647 (AF647) commercially available from a competitor. Whilst in Figure 2 the highest fluorescence signal is seen with the recombinant anti-F4/80 IgG2 (the isotype of the original hybridoma-derived antibody), the isotype control IgG2b (orb256396) shows considerable signal overlap, indicative of binding of the antibody to Fc-receptors. This illustrates the importance of isotype controls in such experiments when using conventional antibody formats particularly when Fc-blocking reagents are incompatible with the system used due to reactivity with the secondary antibody. The Fc silent™ format however overcomes this issue as seen in Figure 3, where the Fc silent™ recombinant anti-F4/80 yields a strong and distinct signal, whilst the isotype control shows no discernible difference to the background staining from the secondary antibody alone. Therefore, with Fc Silent™ reagents, no Fc-blocking products are required.

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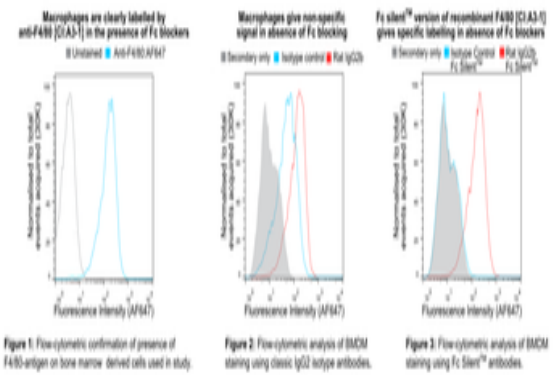

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