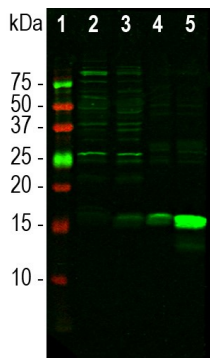


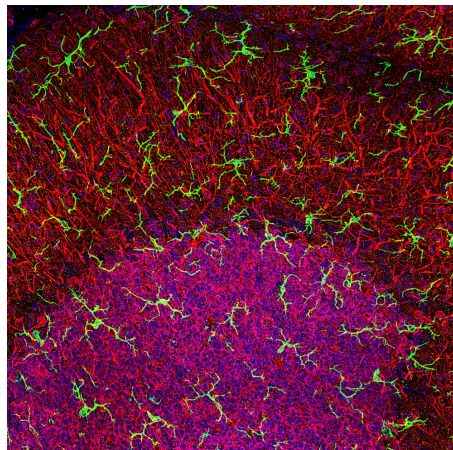
|                          |   |
|--------------------------|---|
| <b>Product name:</b>     | IBA1 Rabbit Monoclonal Antibody   |
| <b>Cat number:</b>       | MAB-94785   |
| <b>Conjugate:</b>        | Unconjugated  |
| <b>Size:</b>             | 100 ul  |
| <b>Clone:</b>            | EPR16589  |
| <b>Concentration:</b>    | 1mg/ml  |
| <b>Host:</b>             | Rabbit  |
| <b>Isotype:</b>          | IgG   |
| <b>Immunogen:</b>        | Peptide identical to the C-terminal of human IBA1 coupled to KLH  |
| <b>Reactivity:</b>       | Hu, Ms, Rt  |
| <b>Applications:</b>     | Western Blot: 1:1,000-2,000. Immunofluorescence:1:1,000-2,000<br>Immunocytochemistry: 1:1,000-2,000 Immunohistochemistry(paraffin-tissues):<br>1:1000 Immunohistochemistry (frozen-tissues): 1:1000 "Free floating" |
| <b>Molecular Weight:</b> | 17kDa   |
| <b>Purification:</b>     | Aff.Pur.  |
| <b>Form:</b>             | Liquid  |
| <b>Buffer:</b>           | Supplied as an aliquot of serum plus 5mM NaN3   |
| <b>Storage:</b>          | Stable at 4°C for one year, for longer term store at -20°C. Avoid freeze/thaw cycles.   |

**Background:**

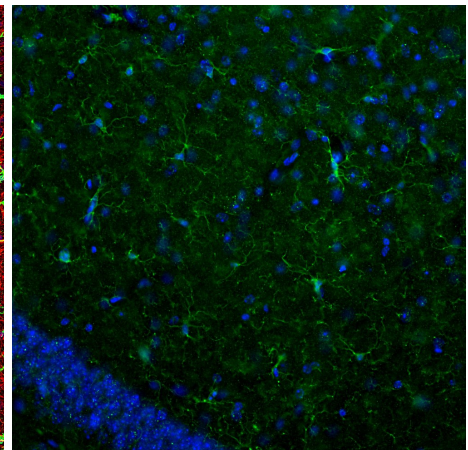
IBA1 is an acronym for “ionized calcium binding adapter molecule 1”, and the protein is also known as AIF1 for “allograft inflammatory factor 1”. AIF1 was originally identified, cloned and sequenced as a protein heavily upregulated in an animal model of graft rejection (1). The AIF1 protein was localized in macrophages and neutrophils surrounding and infiltrating the graft site. Shortly afterwards the same protein was identified as IBA1 in a screen for cytokine induced genes in neurons (2). In the event the workers identified a gene product which was neither expressed in neurons nor induced by cytokines, but which had some very interesting properties, including the important observation that IBA1 was only expressed in hematopoietic cells. IBA1 and AIF1 were subsequently found to be identical, being a small globular 17kDa molecule belonging to the “EF” hand superfamily of Calcium binding proteins. As with other related molecules IBA1 probably has a role in Calcium buffering and in the responses of cells to changes in the level of cellular Calcium. IBA1 is specifically expressed in hematopoietic cells such as neutrophils, macrophages and monocytes. Since the only hematopoietic cells normally found within the central nervous system are microglia, suitable IBA1 antibodies are widely used to identify microglial cells in sections and tissues. Microglia are the immunocompetent cells of the CNS and are extremely important in responses to injury and disease. Microglial are small but very active cells which constantly send processes probing their neighborhood and which alter morphology and are induced to divide following a variety of CNS compromises.



Western blot analysis of different tissue lysates using rabbit mAb to IBA1, IBA1, dilution 1:1,000 in green: [1] protein standard (red), [2] mouse brain, [3] rat brain, [4] mouse spleen, and [5] rat spleen. The band at about 15kDa mark corresponds to IBA1 protein. IBA1 is a relatively minor protein of brain and is much more abundant in spleen, so the 15kDa band is less obvious in CNS lysates. The other bands seen in the CNS lysates are of unknown origin but do not appear to compromise the microglial specific staining seen with this antibody.



High magnification stacked confocal image of rat cerebellar molecular layer at top and granular layer below, stained with IBA1, dilution 1:1,000, in green. Microglia are very small cells with fine processes spreading in three dimensions and so are best visualized in a confocal Z-stack. Red shows the processes of Purkinje cells and the perikarya of granule cells revealed with an antibody to MAP2. Nuclear DNA is shown with DAPI stain in blue.



Immunofluorescent analysis of mouse hippocampus section stained with rabbit mAb to Iba1, dilution 1:1,000 in green. The blue is Hoechst staining of nuclear DNA. Following transcardial perfusion of mouse with 4% paraformaldehyde, brain was post fixed for 24 hours, immersed to 15, then 30% sucrose, froze and cut to 45 μm. Free-floating sections were stained with above antibodies. Microglia are very small cells with fine processes spreading in three dimensions.