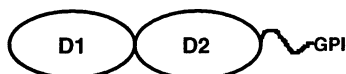


## AS1.2 CD48 cluster report

LLOYD B. KLICKSTEIN and TIMOTHY A. SPRINGER

### CD48 (BLAST-1)

	Epitope 1	Epitope 2	Epitope 3	Epitope 4
Mr 43 kD	WM63 J4-57 J-51	WM68 K31 Tu145	LoMn25	HuLym3



#### Not mapped

MEM-102  
MEM-124  
6.28

CD48 was clustered in the Fourth Workshop [1] on the basis of six monoclonal antibodies (mAb) submitted from five laboratories. There were six CD48 mAb studied in the Fifth Workshop, four of which were submitted to the Adhesion Section, namely, S014 (MEM-102), S018 (MEM-124), S028 (6.28), and S276 (Mo2PT501). The latter was submitted as an mAb that inhibited the allogeneic mixed leucocyte reaction (MLR) and antigen-specific T-cell proliferation and was found by Blind Panel analysis and enzyme-linked immunosorbent assay (ELISA) to recognize CD48. The two CD48 mAb submitted to the T-cell Section, T120 (LOMN25) and T155 (WM68), also were studied previously in the Fourth Workshop. At the time of the Fourth Workshop it was not recognized that CD48 and the BLAST-1 antigen were the same structure [2]. An additional mAb to BLAST-1 was studied in the Fourth Workshop in the B-cell Section.

### Cellular expression

The cellular distribution of CD48 was well established in the Fourth Workshop by flow cytometric and immunohistochemical methods [3]. CD48 expression is restricted to lymphocytes of all subsets, NK cells, and monocytes, with lower levels of expression on eosinophils. Neutrophils are essentially negative but acquire CD48 cell surface antigen upon cytokine activation. Several sarcoma cell lines expressed

low levels of CD48 antigen; however, carcinomas, epithelial cell lines, and purified stromal elements were negative [3; Shaw *et al.*, BP1.3].

### Immunochemistry

CD48 is a 43-kDa glycoprotein initially identified as a B-cell antigen that was upregulated following Epstein-Barr virus (EBV) infection of B lymphocytes [4]. CD48 is a single polypeptide chain that migrates slightly slower after reduction in sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) analysis, suggesting the presence of disulfide bonds. After N-glycosidase F digestion, CD48 migrated at 26 kDa [5]. More recently, CD48 was shown to be attached to the membrane exclusively via a glycosylphosphatidylinositol (GPI) anchor [5,6].

### Molecular cloning

CD48 was first cloned as the B-cell activation marker, BLAST-1 [9]. The extracellular region is comprised of two Ig superfamily domains containing 217 amino acids [7]. CD48 is most homologous to CD58 with 25 per cent sequence identity [5]. The OX45 antigen is probably the rat counterpart of CD48, with 50 per cent sequence identity [5,8]. Murine CD48 is the BCM1 antigen [9]. The CD48 gene is at least 28.6 kb [10] and located on 1q21.3-1q22 [5]. The CD2 and CD58 (LFA-3) genes are also found on chromosome 1, but at 1p13 [11].

### Transfectant and epitope analysis

Two studies addressed the specificity of the CD48 mAb. An ELISA employing soluble recombinant CD48 protein was used to confirm the specificity of Adhesion Section mAb (Ianneli and Thorley-Lawson) and reciprocal mAb cross-blocking studies were performed with the mAb from the T-cell section and non-Workshop CD48 mAb [Henniker and Bradstock, AS1.14]. The latter found four distinct CD48 epitopes:

epitope 1 is recognized by WM63, J4-57, and J-51; epitope 2 is recognized by T155 (WM68), K31, and Tü145; epitope 3 is defined by mAb (T120 (LOMn25)); and epitope 4 is identified by mAb HuLym3 (see introductory diagram).

### Function

There has been no specific function assigned to CD48 in humans, although a recent study in the murine system clearly demonstrated that CD48 is a ligand for CD2 [9]. However, no CD58 counterpart has been identified in rodents thus far and, in contrast to humans, CD48 is found on rodent endothelium [12]. Thus, CD48 in rodents may play the same role as CD58 in humans.

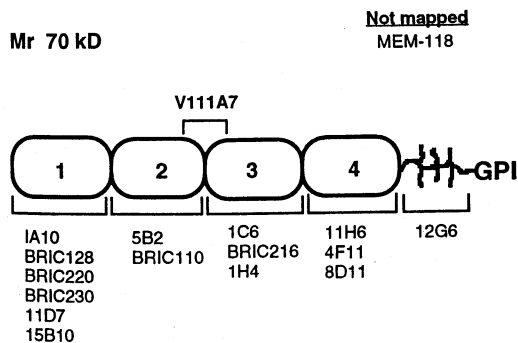
### References

1. Hadam, M. R. In *Leucocyte typing IV* (ed. W. Knapp *et al.*), p. 661. Oxford University Press, Oxford (1989).
2. Yokoyama, S., Staunton, D., Fisher, M., Amiot, J. J., Fortin, J. J., and Thorley-Lawson, D. A. *J. Immunol.* **146**, 2192 (1991).
3. Henniker, A. J., Bradstock, K. F., and Atkinson, K. In *Leucocyte typing IV* (ed. W. Knapp *et al.*), p. 664. Oxford University Press, Oxford (1989).
4. Thorley-Lawson, D. A., Schooley, R. T., Bhan, A. K., and Nadler, L. M. *Cell* **30**, 415 (1982).
5. Staunton, D. E., Fisher, R. C., LeBeau, M. M., Lawrence, J. B., Barton, D. E., Francke, U., Dustin, M., and Thorley-Lawson, D. A. *J. exp. Med.* **169**, 1087 (1989).
6. Hollander, N., Low, M. G., and Springer, T. A. In *Leucocyte typing IV* (ed. W. Knapp *et al.*), p. 182. Oxford University Press, Oxford (1989).
7. Staunton, D. E. and Thorley-Lawson, D. A. *EMBO J.* **6**, 3695 (1987).
8. Killeen, N., Moessner, R., Arvieux, J., Willis, A., and Williams, A. F. *EMBO J.* **7**, 3087 (1988).
9. Kato, K., Koyanagi, M., Okada, H., Takanashi, T., Wong, Y. W., Williams, A. F., Okumura, K., and Yagita, H. *J. exp. Med.* **176**, 1241 (1993).
10. Fisher, R. C. and Thorley-Lawson, D. A. *Mol. cell. Biol.* **11**, 1614 (1991).
11. Sewell, W. A., Palmer, R. W., Spurr, N. K., Sheer, D., Brown, M. H., Bell, Y., and Crumpton, M. J. *Immunogenetics* **26**, 276 (1988).
12. Wong, Y. W., Williams, A. F., Kingsmore, S. F., and Seldin, M. F. *J. exp. Med.* **171**, 2115 (1990).

## AS1.3 CD55 cluster report

LLOYD B. KLICKSTEIN and TIMOTHY A. SPRINGER

### CD55 (DAF)



CD55 (decay accelerating factor, DAF) was clustered in the Fourth Workshop [1] with four monoclonal antibodies (mAb). Three CD55 mAb were studied in the Fifth Workshop, S016 (MEM-118) and S031 (IA10) in the Adhesion Section and T158 (143-30) in the T-cell section.

### Cellular expression

CD55 is widely distributed. Previous reports described the absence of CD55 on NK cells [2]; however, a recent study with a more sensitive flow cytometer found low levels of CD55 on CD56+ cells [Solomon *et al.*, AS1.12]. Studies in this Workshop confirmed previous studies [1] demonstrating the absence of CD55 antigen on the surface of affected cells of