

AS

[Klickstein and Springer, AS1.1, Table 1], defining two to three epitopes (see introductory diagram).

Function

An extensive literature supports the identification of CD59 as an inhibitor of complement-mediated lysis. CD59 acts to inhibit the C5b-8 catalysed insertion of C9 into the lipid bilayer [8,9]. *In vitro* studies have demonstrated binding of purified CD59 protein to purified C8 and C9 [10]. CD59 is highly expressed on all haemopoietic cells and mAb to CD59 as well as to CD48 on lymphocytes were shown to be the most sensitive and specific for diagnosis of paroxysmal nocturnal haemoglobinuria (PNH) (Alfinito) by flow cytometry. Genetic deficiencies in CD59 [11] or CD55 [12,13] have shown that the CD59-deficient patient had clinical PNH, while the CD55 patients were apparently normal. Although the number of patients studied was small, CD59 is apparently the most important inhibitor of complement-mediated lysis on the erythrocyte surface.

Cells transfected with CD2 and purified CD59 protein have been used to demonstrate that CD59 is also a counterreceptor for CD2 [14].

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AS1.6 CDw108 cluster report

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Two monoclonal antibodies (mAb), S015 (MEM-121) and S017 (MEM-150), both IgM from Hořejší's laboratory but derived from different fusions, defined the CDw108 cluster in this Workshop.

Cellular distribution

CDw108 is expressed highly only on HPB-ALL cells, and weakly on some lymphoid, myeloid, and stromal cells. One study found CDw108 to be regulated during B-cell development, with B-cell chronic lymphocytic

leukaemia (B-CLL) cells negative, Epstein-Barr virus (EBV)-transformed B cells and hairy leukaemia cells positive, and myelomas and plasmacytomas negative (Halperin).

Immunochemistry

CDw108 mAb immunoprecipitate and detect a 75 kDa band in Western blots of non-reduced sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) gels [Angelisová *et al.*, AS1.8]. The

The band is 80 kDa after reduction. The antigen contains about 20 per cent of N-linked carbohydrate as determined by endoglycosidase F treatment. The antigen identified by both CDw108 mAb is 100 per cent glycosylphosphatidylinositol (GPI)-anchored on HPB-ALL [Angelisová *et al.*, AS1.8] and JY cells [Klickstein *et al.*, AS1.7].

No specific functions were identified during the course of this Workshop; however, many GPI-anchored antigens have been shown to be associated

with protein tyrosine kinases [Angelisová *et al.*, AS1.8] or to mediate cellular activation or proliferation of some cells. In HPB-ALL, CDw108 antigen is associated with several other GPI-linked molecules, glycolipids, and with protein tyrosine kinases [Angelisová *et al.*, AS1.8]. It is clear that mAb to CDw108 do not affect T-cell proliferation or human erythrocyte E^{hu} rosetting in the assays studied in this Workshop [Klickstein and Springer, AS1.1, Table 2].

AS1.7 Identification of novel GPI-anchored antigens by analysis of GPI-anchor-deficient cells with mAb in the Blind Panel

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The glycosylphosphatidylinositol (GPI) anchor [1] is a posttranslational modification of a nascent polypeptide chain where a characteristic COOH-terminal signal peptide directs the concerted proteolytic cleavage and transfer of a GPI moiety [2-4]. The GPI-anchored protein is then transported via the endoplasmic reticulum to the cell membrane where the long-chain fatty acids of the GPI group attach the protein to the outer leaflet of the lipid bilayer. Some of the enzymes in the GPI biosynthetic pathway have been cloned [5].

To identify novel GPI-anchored antigens, clones of a B lymphoblastoid cell line, an erythroleukaemia cell line, and a fetal kidney epithelial cell line (Table 1) were analysed by indirect immunofluorescence and flow cytometry with the 480 monoclonal antibodies (mAb) of the Fifth Workshop Blind Panel and with the 31 mAb in Subpanel 1 of the Adhesion Section. The staining pattern of the normal phenotype clone was compared to that obtained with corresponding GPI-anchor-deficient cells and with that of the

Table 1 Established GPI-anchored antigens

CD	Antigen	Workshop mAb		Cell line*	% loss of staining on GPI-anchor-deficient clone
		Code	Clone name		
CD24	HSA, B7-2	CD24.3	HB-8	K-562	100
CD48	BLAST-1	S028	6.28	JY	100
CD52	CAMPATH-1	XB003	O97	K-562, JY	100
CD55	DAF	S031	IA10	K-562, JY, 293	100
CD58	LFA-3	S024	TS2/9	K-562, JY	50
CD59	MIRL	S013	MEM-43	K-562, JY, 293	100
CD14	P55-65	MR3	MoS39		
CD16	Fc γ RII	MR5	3G8		
CD56	NCAM	NK19	Leu 19		
CD67	CGM6	M40	MF25.1		
CD73	Ecto 5'NT	CD73.1	1E9.28.1		

*Cell line in which mAb stained normal cell clone but not the GPI-anchor-deficient clone.

†% of staining of normal cell clone absent on GPI-anchor-deficient clone.