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Immunology

CD22 associates with the human surface IgM–B-cell antigen receptor complex

(membrane immunoglobulin/B lymphocyte/signal transduction/phosphorylation)

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ABSTRACT **The B-cell surface molecule CD22, when cross-linked, modulates signaling through the surface IgM (sIgM)–B-cell receptor (BCR) complex. Here we analyzed the basis of this interaction between CD22 and the human sIgM complex. After lysis of B cells or B-cell lines in digitonin, CD22 coimmunoprecipitated a kinase activity that *in vitro*-phosphorylated two polypeptides of 150 and 130 kDa on tyrosine residues. By immunoblot analysis with a rabbit antiserum specific for a synthetic peptide of CD22, we found these proteins to be CD22 itself. Furthermore, the phosphorylated 150-kDa CD22 was found in the sIgM–BCR complex maintained by digitonin, along with Igα/mb-1, Igβ/B29, and a 75-kDa polypeptide precipitated by an antiserum specific to protein-tyrosine kinase PTK72. CD22 is likely to be an important signaling partner in the sIgM–BCR complex since it is very rapidly and strikingly phosphorylated after sIgM is cross-linked and since it contains the antigen recognition homology I (ARHI) motif, present in other antigen receptor molecules.**

CD22 is a B-cell-restricted marker expressed in the cytoplasm of pre-B cells and on the surface of mature B lymphocytes (1, 2). Isolation and sequencing of cDNAs encoding for human and murine CD22 have revealed that CD22 is a member of the immunoglobulin superfamily (3–5). CD22 is an adhesion molecule (3–5); it binds CD45RO, an isoform of the CD45 protein-tyrosine phosphatase expressed on a subset of T lymphocytes (6, 7), and it may bind other ligands (3, 4, 6). Previously, it was found that cross-linking CD22 with monoclonal antibodies (mAbs) potentiated B-cell activation induced by anti-IgM and that only $CD22^+$ lymphocytes showed an increase in intracellular free calcium upon cross-linking the surface IgM (sIgM)–B-cell receptor (BCR) complex (8, 9). We suggested that CD22 may be an essential bridge molecule for transducing sIgM-dependent signals and that CD22 may interact functionally with the sIgM–BCR complex.

The structure of the sIgM–BCR complex has been elucidated and it has similarities with the CD3–T-cell antigen receptor (TCR) complex (10–17). The sIgM receptor associates with at least two other membrane polypeptides, Igα and Igβ, encoded by the *mb-1* (18) and *B29* (19) genes, respectively. Intracellular protein-tyrosine kinases (PTKs) including the Src-family PTK (20–23) and the PTK72 PTK (24) also interact with the sIgM–BCR complex. When analyzing human B cells, we found that sIgM associates not only with human Igα and Igβ but also with two unidentified polypeptides of 75 kDa and 150 kDa that are phosphorylated after sIgM cross-linking (23).

Here we report that the 150-kDa component associated with the human IgM–BCR complex is CD22. To better understand how the sIgM-associated elements participate in signal transduction, we analyzed the signal generated by CD22 and its connection to the sIgM pathway. We found that surface CD22 physically interacts with a kinase activity that can phosphorylate CD22 *in vitro*, mainly on tyrosine residues. CD22 was identified in the sIgM–BCR complex maintained by digitonin along with Igα, Igβ, and the human 75-kDa B-cell equivalent of mouse PTK72/Syk. Based on functional and structural data, CD22 is likely to work in close association with the sIgM–BCR multiprotein signaling complex.

MATERIALS AND METHODS

Cells. The Ramos and Daudi Burkitt lymphoma lines and tonsillar B cells (E-rosette-negative, >90% $CD20^+$) were prepared as described (25). B cells were separated on discontinuous Percoll gradients (Pharmacia), and cells at the 60–50% (vol/vol) Percoll interface were used for these studies.

Reagents. Purified anti-human μ chain (4B8, IgG2a), CD22 (HD39 and G28-7, IgG1), and CD19 (HD37, IgG1) were used for immunoprecipitations (26–28). MOPC21 (IgG1) and a CD8 mAb (G10-1, IgG2a) were used as isotype-matched control mAbs. For Western blot experiments, purified rabbit anti-phosphotyrosine sera were prepared as described (29). Rabbit antisera to PTK72 were produced as described (30). Rabbit antisera were also generated against two synthetic peptides of the intracytoplasmic tail of human CD22 coupled to keyhole limpet hemocyanin (peptide 1, RNKKVRRAPLSEGPH; peptide 2, EMNIPRTGDAESSEM). Both of these antisera reacted equally well and specifically with immunoprecipitated CD22.

Cell Surface Labeling with ^{125}I. B cells were suspended in 0.5 ml of phosphate-buffered saline (PBS) and incubated for 10 min at room temperature with 1 mCi of $Na^{125}I$ (100 mCi/ml; 1 Ci = 37 GBq; ICN), 30 μg of lactoperoxidase (100 units/mg, Sigma), and 100 μl of H_2O_2 (0.03% in PBS). Cells were washed once in an ice-cold solution of 150 mM NaI in PBS and twice in ice-cold RPMI 1640 medium containing 10% (vol/vol) fetal calf serum and then were lysed either in 50 mM Tris·HCl/150 mM NaCl/5 mM EDTA, pH 8.0, containing 0.5% Nonidet P-40 (NP-40) or in 10 mM triethanolamine/150 mM NaCl, pH 7.8, containing 1.0% digitonin. In both cases, the lysis buffer contained protease inhibitors [1 mM phenylmethylsulfonyl fluoride/aprotinin (1 μg/ml)/leupeptin (1 μg/

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Abbreviations: ARHI, antigen recognition homology I; BCR, B-cell antigen receptor; FcR, receptor for the Fc portion of immunoglobulin; mAb, monoclonal antibody; PTK, protein-tyrosine kinase; sIgM, surface IgM; TCR, T-cell antigen receptor; NP-40, Nonidet P-40.

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ml)/soybean trypsin inhibitor (100 μg/ml)] and 2 mM sodium orthovanadate (Sigma), a phosphatase inhibitor. After lysis, the samples were centrifuged at 15,000 × *g* for 30 min.

Immunoprecipitation, SDS/PAGE, and *in Vitro* Kinase Assays. Cell lysates were incubated for 3 h with soluble antibody followed by a 1-h incubation with protein A or protein G beads (Pharmacia). For digitonin lysates, beads were washed five times in triethanolamine/NaCl, pH 7.8, containing 0.2% digitonin. For NP-40 lysates, beads were washed five times as described (23). Unless indicated, beads were resuspended in SDS/PAGE sample buffer and boiled prior to electrophoresis on 10% polyacrylamide gels. The *in vitro* kinase assay was performed as described (23).

Phosphoamino Acid Analysis. The phosphoamino acid analysis of phosphorylated proteins was performed as described (23, 31). Bands were excised from dried gels and rehydrated. Proteins were precipitated with trichloroacetic acid before dilution in 50 μl of constantly boiling HCl (5.7 M, Sigma) and heating for 1 h at 110°C in a N_2 atmosphere. Samples were then applied onto a TLC plate for two-dimensional electrophoresis. Dry plates were sprayed with ninhydrin solution and heated briefly at 80°C to develop the standard amino acids before autoradiography.

Western Blot Analysis. Proteins separated by SDS/PAGE were transferred to an Immobilon membrane (Millipore). After saturation in 10 mM Tris·HCl/150 mM NaCl, pH 7.2, containing 5% (wt/vol) bovine serum albumin and 20% fetal calf serum, the immunoblots were incubated for 3 h with anti-phosphotyrosine antibodies (0.5 μg/ml) as described (29) or with rabbit anti-CD22 antiserum (1:1000 dilution). Thereafter, membranes were washed and antibody binding was detected with high-specific-activity ^{125}I-labeled protein A (ICN; 1 μCi/ml).

RESULTS

CD22 Is Associated with a Protein Kinase Activity. To test whether CD22 was associated with a protein kinase activity, we immunoprecipitated CD22 after lysis of B-lineage cells in the mild detergent digitonin. We then tested CD22 immunoprecipitates in an *in vitro* kinase assay. CD22 from two B-cell lines and from normal tonsillar B cells contained a phosphorylated doublet of 150/130-kDa polypeptides, the larger protein being more heavily labeled (Fig. 1*A*). A phosphoamino acid analysis (Fig. 1*B*) revealed that the 150/130-kDa proteins were phosphorylated mainly on tyrosine and to a lesser extent on serine residues, suggesting that CD22 associates with a tyrosine kinase and possibly a serine kinase active in an *in vitro* assay. When the immunoprecipitating antibody was incubated with viable cells before lysis, instead of being added to the whole lysate, the same phosphorylated polypeptides were detected after an *in vitro* kinase assay, indicating that the membrane form of CD22 associates with a kinase activity (data not shown). Furthermore, kinase assays performed on CD22 from NP-40 lysates did not show any phosphorylated proteins (data not shown). This suggests that CD22 is associated with a kinase via an interaction disrupted by NP-40. In Western blot analysis, 150- and 130-kDa proteins were recognized by an antiserum generated against a synthetic peptide of the cytoplasmic tail of CD22, suggesting that the phosphorylated bands were in fact CD22 (Fig. 1*C*).

CD22 Can Associate with sIgM. The phosphorylated CD22 has the same electrophoretic mobility as a 150-kDa component of the human sIgM complex (23). This 150-kDa polypeptide is phosphorylated during an *in vitro* kinase assay performed on IgM immunoprecipitates (Fig. 2*A*). This suggested that CD22 might interact with sIgM, a hypothesis reinforced by ^{125}I-labeling experiments (Fig. 2*B*). ^{125}I-labeled and digitonin-solubilized CD22 was immunoprecipitated with slightly labeled bands of ≈30 kDa and ≈80 kDa, similar to the molecular masses of the light and heavy chains of IgM, respectively. These two bands were not found in NP-40-solubilized CD22 immunoprecipitates, suggesting that NP-40 disrupts the interaction between CD22 and the putative sIgM (Fig. 2*C*). Our inability to detect a 150-kDa polypeptide in IgM immunoprecipitates from an ^{125}I-labeled cell lysate could reflect the stochiometry of the interaction of CD22 and sIgM and suggests that only a small fraction of surface CD22 associates with sIgM. The two CD22 mAbs used, HD39 and G28-7, recognize distinct epitopes on CD22 (1, 27), which may account for the different immunoprecipitation patterns

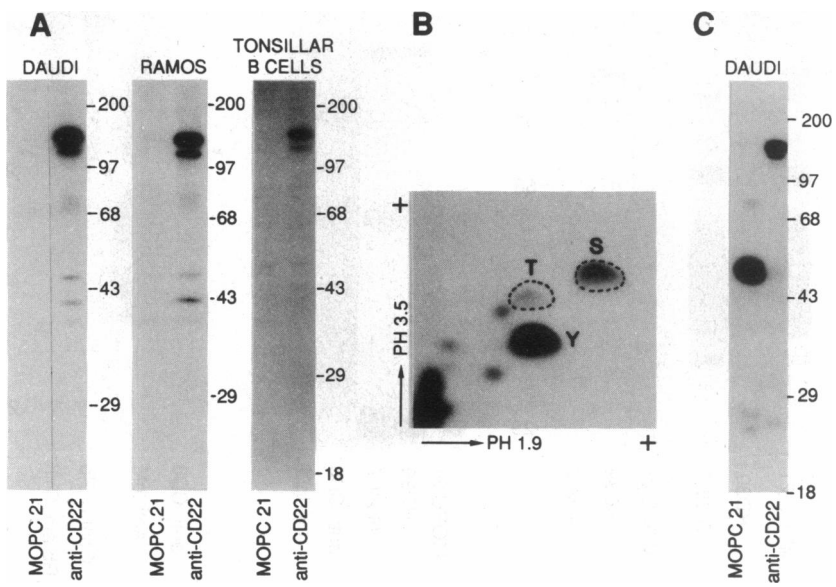

FIG. 1. CD22 associates with a protein kinase activity. (*A*) CD22 was immunoprecipitated with HD39 mAb from digitonin-lysed Daudi, Ramos, or tonsillar B cells (negative control, MOPC21 cells). *In vitro* kinase assays with [γ-^{32}P]ATP were performed on the immunoprecipitates before SDS/PAGE. (*B*) ^{32}P-labeled CD22 from Daudi cells was analyzed for phosphoamino acid content. Positions of standard amino acids are shown by dotted lines (Y, phosphotyrosine; S, phosphoserine; T, phosphothreonine). (*C*) A CD22 immunoprecipitate (or MOPC21 control) from digitonin-lysed Daudi cells was separated by SDS/PAGE, transferred onto a membrane, and blotted with a rabbit anti-CD22 serum. The band at ≈50 kDa in the control lane is due to the interaction of ^{125}I-labeled protein A with the MOPC21 heavy chain. Molecular masses (kDa) are in the margins.

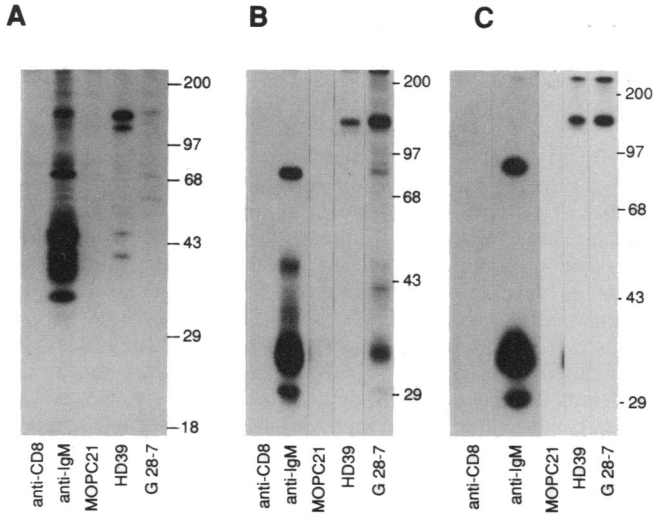

FIG. 2. CD22 at 150 kDa is the same size as one of the components of the sIgM complex. (*A*) Digitonin-lysed Daudi cells were used to immunoprecipitate IgM (4B8 mAb) or CD22 (HD39 or G28-7 mAb). MOPC21 and G10-1/CD8 mAbs were negative controls. *In vitro* kinase assays with [γ-^{32}P]ATP were performed on the immunoprecipitates. Autoradiography was performed for 30 min. (*B*) Daudi cells were ^{125}I-labeled and lysed in digitonin. The two left lanes were exposed for 8 h; the three right lanes were exposed for 48 h. (*C*) Daudi cells were ^{125}I-labeled and lysed in NP-40. The two left lanes were exposed for 8 h; the three right lanes were exposed for 24 h. The immunoprecipitations in *B* and *C* were performed with the same antibody used in *A*. Molecular masses (kDa) are indicated.

obtained with the two mAbs when different labeling conditions were used.

To examine further the structural relation between sIgM and CD22, we performed sequential immunoprecipitations. First, the sIgM complex was isolated from Daudi cells and phosphorylated *in vitro* (23). The kinase reaction was stopped by the addition of NP-40 buffer, which also disrupted the multiprotein complex. The supernatant was used for a second immunoprecipitation with CD22 mAb (Fig. 3), which specifically removed the 150-kDa polypeptide from the pool of sIgM complex elements (Fig. 3*A*) and secondarily immunoprecipitated a 150-kDa protein not present in the control lanes (Fig. 3*B*). The same results were obtained with normal tonsillar B cells (data not shown). A rabbit antiserum specific for peptides from the cytoplasmic tail of CD22 that recognizes immunoprecipitated CD22 when used in immunoblot analyses also reacted with the 150-kDa band of the sIgM complex (Fig. 3*C*). Thus, these results demonstrate that the B-cell-specific CD22 molecule can associate with the sIgM complex. Furthermore, the phosphorylated 75-kDa polypeptide in the IgM complex could be precipitated by a specific anti-PTK72 antibody (Fig. 3*D*) and apparently is the human B-cell equivalent of mouse PTK72 kinase (30).

Functional Interaction Between sIgM and CD22. Pretreatment of B cells with CD22 mAb potentiates their ability to respond to an anti-IgM-induced signal (8, 9). Thus, we examined whether the signaling pathways of sIgM and CD22 were closely connected. Using the same conditions previously used for measurements of intracellular free calcium, we found that pretreatment of viable B cells with CD22 mAb did not alter the phosphotyrosine content of an IgM immunoprecipitate (data not shown). However, when Daudi cells were stimulated for 0.5, 5, or 30 min with either CD22 mAb or anti-IgM mAb before lysis in NP-40 and immunoprecipitation of CD22, the results were different (Fig. 4). After SDS/PAGE, proteins were detected by Western blot analysis with anti-phosphotyrosine antibody (Fig. 4*A*). Stimulation of B cells with anti-IgM mAb dramatically increased the phosphotyrosine level of CD22 as early as 30 s after cross-linking, whereas CD22 stimulation had a small but significant effect. Western blot analysis with anti-CD22 showed no major variation in the actual amount of CD22 immunoprecipitated (Fig. 4*B*). Thus, in viable B cells, sIgM and CD22 do communicate, as sIgM cross-linking signals a rapid change in

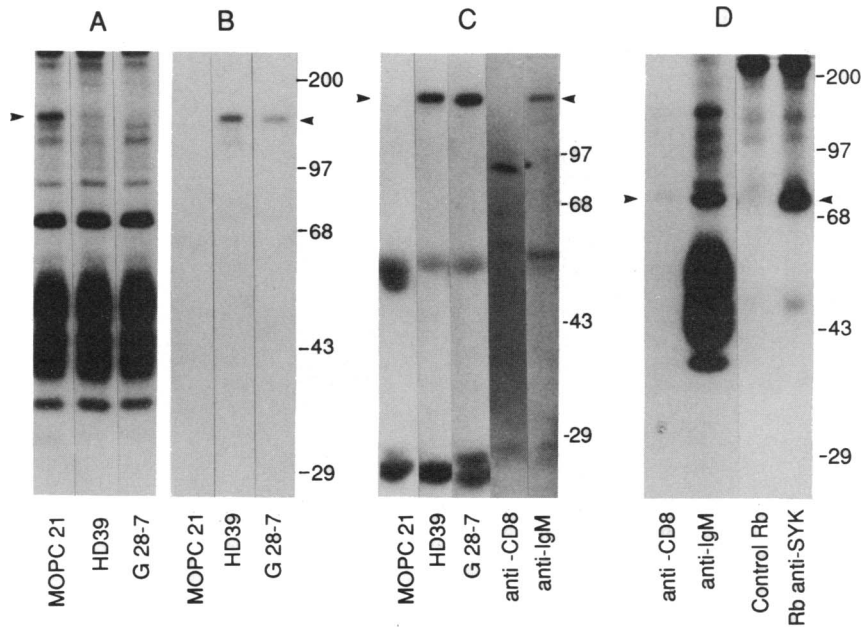

FIG. 3. CD22 associates with the sIgM–BCR complex. (*A*, *B*, and *D*) Digitonin-lysed Daudi cells were used for immunoprecipitation of IgM. *In vitro* kinase assays were performed on IgM immunoprecipitates. Kinase reactions and molecular associations were stopped by adding buffer containing NP-40. (*A* and *B*) Supernatants were submitted to second-step immunoprecipitations with CD22 (HD39 or G28-7) or control (MOPC21) mAb. The leftover supernatants from the second step are shown in *A* (film exposed for 30 min) and the second-step immunoprecipitates are shown in *B* (film exposed 1 h). (*D*) Supernatant from the IgM immunoprecipitate (second lane) was reimmunoprecipitated with anti-PTK72 or a control rabbit antiserum. (*C*) CD22 (HD39 and G28-7 mAbs), IgM (4B8 mAb), or control immunoprecipitates from digitonin-lysed Daudi cells were separated by SDS/PAGE and analyzed on a Western blot with anti-CD22 antiserum. Standard molecular masses (kDa) are indicated.

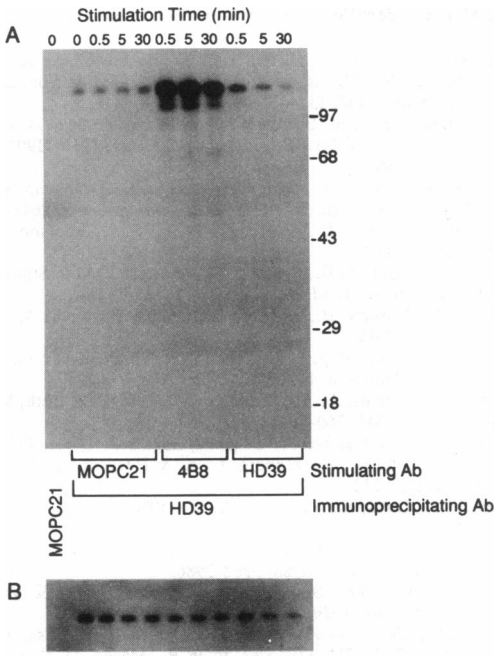

FIG. 4. CD22 is phosphorylated on tyrosine after cross-linking sIgM. Daudi cells were incubated at 37°C for 0.5, 5, or 30 min with CD22 (HD39), anti-IgM (4B8), or control (MOPC21) mAb and lysed in NP-40. CD22 was immunoprecipitated from each of these samples, divided in half for SDS/PAGE, and then analyzed on a Western blot with either anti-phosphotyrosine antibody (*A*) or anti-CD22 antiserum (*B*). Standard molecular masses (kDa) are indicated. Ab, antibody.

phosphotyrosine content of CD22. Similar results were recently reported by Schulte *et al.* (32).

Comparison Between Cytoplasmic Tail Sequence of CD22 and Other Receptor Molecules. A particular structural motif—antigen recognition homology I (ARHI) (33)—is present in CD3-TCR and sIgM-BCR molecules and in certain Fc receptor (FcR) chains. We used the PILE-UP/ALIGNMENT program to compare the intracytoplasmic amino acid sequences of human and murine CD22 with these receptors. The CD22 sequences contain six tyrosine residues, of which two are organized in a typical ARHI-like motif—(D/E)-XXX(Y)XX(L/I)XXXXXXX(Y)XX(V). The number of residues between the negatively charged amino acid (E or D) and the C-terminal tyrosine is different from the standard ARHI motif, which is also true for the cytoplasmic tail of FcγRII/CD32 (Fig. 5). Two CD22 tyrosine residues (Y822 and Y842) are separated by 19 amino acids and thus are not organized in a strict ARHI motif; however, they may still function as an ARHI motif, as the ARHI motif of CD32 presumably does even though it too has a long interval (16 residues). Thus, CD22 may have two functional ARHI motifs; the only antigen receptor chain with more than one ARHI motif is the TCR ζ chain (three motifs).

DISCUSSION

Here we show that CD22 can associate with the human sIgM–BCR complex and is likely to participate in sIgM-dependent signaling. After lysis of B cells with a mild nonionic detergent, CD22 is immunoprecipitated in association with a kinase activity that can *in vitro*-phosphorylate 150- and 130-kDa polypeptides, mainly on tyrosine residues. The 150- and 130-kDa proteins were identified as CD22 itself by a combination of immunoprecipitation and immunoblot experiments using CD22 mAbs and polyclonal antisera to CD22 peptides. CD22 was described (34, 35) as a doublet of ≈130/140 kDa. The difference in the size of CD22 in these reports and our studies is probably due to small experimental variations. The heavily phosphorylated 150-kDa CD22 molecule was subsequently shown to be the same size as a component associated with the sIgM complex (23). Both two-step immunoprecipitations and Western blot analysis established that CD22 is indeed the molecule associated with the BCR complex maintained by digitonin.

The regulation and stochiometry of the CD22 association with the sIgM–BCR complex are important issues not yet fully understood. Several results indicate, however, that sIgM-associated CD22 represents a small fraction of the total pool of CD22. (*i*) A pool of isolated CD22 coexists with a pool of sIgM-associated CD22. Comparisons by densitometry of immunoblots of CD22 in sIgM immunoprecipitates vs. immunoprecipitated CD22 suggest that CD22 associated with sIgM is <5% of cellular CD22 (data not shown). (*ii*) Little or no ^{125}I-labeled CD22 was detectable in sIgM immunoprecipitates (Fig. 2*B*); a more sensitive labeling method was needed to detect sIgM-associated CD22 (Fig. 2*A*). (*iii*) sIgM and CD22 do not obviously cocap (data not shown).

Since CD22 can also function as a cell–cell interaction molecule, it is tempting to speculate that binding of CD22 to an external ligand such as CD45RO (6, 7) or another ligand (3, 4, 6) may induce or facilitate the interaction of CD22 with a mobile sIgM–BCR complex and, thereby, potentiate antigen-specific signaling of B cells. The macrophage/dendritic cells present in splenic marginal zone are potential candidates for CD22-ligand-bearing cells (36). Another intriguing possibility is that certain bacteria may express structures able to be bound by CD22, which in turn could facilitate B-cell responses less dependent on T cells. Ligation of CD22 may also increase receptor signals via one of its ARHI motifs (Fig. 5).

The structural association of CD22 and sIgM was reinforced by the finding of a rapidly induced signal between

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HuCD22cyto  752   YNPMMEDGIS YTTLRFPEMN IPRTGDAESS EMQRPPRTCD DTVTYSALHK RQVGDYENVI ....PDFPED
MuCD22cyto  773   ---A-D-TV- -AI----SD THNA---GTP AT-A--PNNS -----VTQ- -PM-------N ....-SC---
Humb-1cyto  166   .......... .......... RKRWQNEKLG LDAGDEYEDE -ENL-EG-NL DDCSM--DIS RGLQGTYQDV
 HuB29cyto  182   .......... .......... ..... DKDSSKAGME EDHT-EG-DI DQTAT--DIV TLRTGEVKWS
HuCD3ζcyto   52   .......... .......... ....RVKF-R SAEP-AYQQG QNQL-NE-NL GRREE-DVLD KRRGRDPEMG
HuCD3δcyto  128   .......... .......... ..HETGRL-G AADTQALLR. NDQV-QP-RD -DDAQ-SHLG GNWARNK...
HuCD3γcyto  138   .......... .......... .GQD-VRQ-R ASDKQTLLP. NQQL-QP-KD -EDDQ-SHLQ GNQLRRN...
HuFcγ2cyto  235   .......... .......YCR KK-ISANSTD PVKAAQFEPP GRQMIAIRKR QLEETNNDYE TADGGYMTLN

HuCD22cyto      EGIHYSELIQ FGVGERPQAQ ENVDYVILKH .......... .......... .......... ...... 847
MuCD22cyto      -S---L--V- --A-K----- -D----T--- .......... .......... .......... ...... 868
Humb-1cyto      GSLNIGD.V- LEKP...... .......... .......... .......... .......... ...... 226
 HuB29cyto      VGEHPGQE .......... .......... .......... .......... .......... ...... 229
HuCD3ζcyto      GKPRRKNPQE GLYNELQKDK MAEA-SEIGM KGERRRGKGH DGLYQGLSTA TKDTYDALHM QALPPR 164
HuCD3δcyto      .......... .......... .......... .......... .......... .......... ...... 172
HuCD3γcyto      .......... .......... .......... .......... .......... .......... ...... 182
HuFcγ2cyto      PRAPTDDDKN IYLTLPPNDH VNSNN..... .......... .......... .......... ...... 311
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FIG. 5. Intracytoplasmic tail of CD22 contains the ARHI motif. The intracytoplasmic portion of human CD22 was compared to murine CD22 and other human receptor molecules for all or part of their cytoplasmic tails. Dashes indicate identities with the human CD22 sequence. Noticeable homology motifs between the sequences are boxed. Hu, human; Mu, murine.

these molecules. It has been suggested (9) that CD22 might be required for efficient signaling via the sIgM–BCR complex, as only CD22$^+$ B cells could be induced to release intracellular calcium upon sIgM cross-linking. Cross-linking sIgM on viable B cells induced tyrosine phosphorylation of CD22 [and also of Igα and Igβ (37)] within seconds (Fig. 4 and ref. 32). Thus, CD22 is likely to be an important partner of the sIgM complex that may be acting downstream of sIgM in the signaling process. Upon sIgM cross-linking, the phosphorylation of CD22 may affect its ability to interact with key elements in the signaling pathway.

To understand how CD22 functions, it will be necessary to determine with which kinase it associates and what regulates this association. The mouse and human sIgM–BCR complexes associate with Src-family kinases including Lyn, Fyn, Blk, and Lck (20–23). Recently, Cambier and coworkers (38) reported that the cytoplasmic tail of Igα/mb-1 interacts with Fyn. Moreover, high molecular mass components of the mouse sIgM–BCR complex associate with the PTK72 kinase (24). In human B cells, the sIgM–BCR complex also associates with PTK72 (Fig. 3*D*): the human sIgM complex maintained in digitonin contains a 75-kDa phosphorylated polypeptide that we were able to reimmunoprecipitate with an anti-PTK72 antiserum. The PTK72 kinase appears to be identical to the spleen tyrosine kinase Syk (39), since antipeptide antibodies specific for porcine Syk recognize mouse and human PTK72 (R.L.G., unpublished results). Whether human PTK72/Syk interacts directly with sIgM and/or CD22 remains to be determined.

The identification of CD22 as a signaling molecule is substantiated by the presence in CD22's cytoplasmic tail of one or perhaps two ARHI motifs (33). These motifs are found in other signaling receptor molecules such as TCR or BCR complex molecules or FcR chains and are believed to be critical for signaling (40, 41). The tyrosine residues of one or more of these chains may get phosphorylated after receptor cross-linking and then interact with the SH2 domains of downstream signaling elements. The CD22 intracytoplasmic tail, like the TCR ζ chain, may have more than one potential ARHI motif. The TCR ζ chain and CD22 have distinct influences on surface expression of the T- and B-cell clonotypic receptors, but one testable speculation is that CD22 and ζ chain use related steps for TCR or BCR signal transduction. The 70-kDa ZAP-70 molecule associates with TCR ζ chain after T-cell activation (42) and after tyrosine phosphorylation of the ζ chain via a mechanism dependent on Src-family PTK. ZAP-70 was recently found to be a tyrosine kinase homologous to PTK72/Syk (43). Others also found a 70-kDa tyrosine kinase activity associated with CD3 after TCR activation (44). Cambier (45) has emphasized the similarities between the TCR and BCR complexes: although Igα–Igβ and Igα–Igγ heterodimers are analogous to CD3–ε/γ and CD3–ε/δ heterodimers, respectively, no obvious analogue has been identified for ζ chain. Could CD22 be functioning and be regulated in an analogous way to TCR ζ chain? According to this model, after sIgM cross-linking, a PTK would be activated and phosphorylate CD22. Phosphorylated CD22 would interact with the human B-cell PTK72/Syk or a Syk-like kinase and thereby provide a part of the signals induced via the sIgM–BCR complex.

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