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Ia-LIKE ANTIGENS ON HUMAN T LYMPHOCYTES: RELATIONSHIP TO OTHER SURFACE MARKERS, ROLE IN MIXED LYMPHOCYTE REACTIONS, AND STRUCTURAL PROFILE[1, 2]

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Less than 1% of nylon wool purified human T lymphocytes react with monoclonal antibodies to Ia-like antigens in an indirect rosette microassay. After PHA stimulation, about 70% of T lymphocytes including both Fcγ and Fcμ receptor bearing subpopulations acquire Ia-like antigens having structural properties similar to those of B lymphoid cell-derived Ia-like antigens. The expression of Ia-like antigens on T cells cultured with PHA occurs either by induction or increased synthesis rather than by clonal expansion of an Ia-like antigen-bearing subpopulation present in unstimulated T cell preparations or by T cell uptake of Ia-like antigens shed from Ia-like antigen-bearing cells. The increase in Ia-like antigen-bearing T cells during a 4-day culture with PHA correlated closely with the synthesis of DNA but was contrasted by a decrease in the percentage of Fcμ receptor T cells to undetectable levels by 48 hr in culture. During this time the percentage of cells bearing Fcγ receptors, HLA-A,B,C antigens, and membrane bound Ig did not show any significant change. Blocking studies showed no relationship between newly expressed Ia-like antigens on PHA-activated T cells and receptors for either sheep erythrocytes or the Fc portion of IgG. PHA-activated T cells bearing Ia-like antigens stimulated a unidirectional allogenic MLR that can be blocked by the addition of monoclonal antibodies to Ia-like antigens but not to HLA-A,B,C antigens. These data indicate that T cell Ia-like antigens are structurally and functionally similar to B cell Ia-like antigens and suggest the importance of these antigens to T cell-activated functions.

Although there is a general agreement that the majority of human peripheral blood T lymphocytes do not express Ia-like antigens but may acquire these antigens after stimulation with mitogens, alloantigens, or conditioned media (1–11), there is still controversy concerning the existence of a minor subpopulation of T cells bearing Ia-like antigens and whether T cell activation directly induces Ia-like antigen expression or selectively expands a clone of Ia-like antigen bearing T cells present in the resting T cell population. Thus, some investigators using polyclonal xenoantisera and an indirect immunofluorescence assay have reported that about 4 to 10% of resting peripheral blood T cells bear Ia-like antigens (5, 9, 10). In contrast Evans *et al.* (4) using a polyclonal xenoantiserum and a ^{51}Cr cytotoxicity assay, and Reinherz *et al.* (8) using monoclonal antibodies and the fluorescence-activated cell sorter, have failed to detect Ia-like antigens on resting peripheral blood T cells. Furthermore, data by Ko *et al.* (6) argues in favor of a clonal expansion theory for some of the Ia^+ T cells since treatment of T cells with C and a xenoantiserum to Ia-like antigens reduced the percentage of cells bearing Ia-like antigens after PHA-activation; however, Reinherz *et al.* (8) who exposed T cells to C and monoclonal antibodies to Ia-like antigens were unable to confirm this result. Ia-like antigens on resting T cells, although controversial, have been assessed for their relationship to Fc receptors (11) and for their ability to function in a mixed lymphocyte reaction (10), whereas little such data exist for the Ia-like antigens expressed on activated T cells (2).

In view of the conflicting results in the literature and the scarcity of information about various aspects of the Ia-like antigens of activated T cells, we have 1) tested resting T cells for expression of Ia-like antigens; 2) investigated the question of clonal expansion *versus* Ia-like antigen induction after PHA-activation; 3) assessed the kinetics of expression of Ia-like antigens and other surface markers as well as their spatial relationships on the cell surface of PHA-activated T cells; 4) characterized Ia-like antigens on T cells for their involvement in allogeneic MLR; and 5) defined the molecular profile of T cell derived Ia-like antigens. These experiments have been performed with monoclonal antibodies in order to avoid false positive reactions from contaminant antibodies present at low levels in polyclonal xenoantisera. In addition, highly purified preparations of T cells were used to eliminate the possibility that T cells may bind Ia-like antigens shed from B cells and/or monocytes.

MATERIALS AND METHODS

Human lymphoid cells. Isolation of peripheral blood lymphocytes (PBL)[4] involved Plasmagel sedimentation, Ficoll-Hy-

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[3] Dr. F. Indiveri is a Visiting Investigator from the University of Genova (Italy).

[4] Abbreviations used in this paper: AET, 2-aminoethylisothiouronium bromide; β_2-μ, β_2-microglobulin; ChE, chicken erythrocytes; MbIg, membrane Ig; MLR, mixed lymphocyte reaction; MoAb, monoclonal antibody; PAGE, polyacrylamide gel electrophoresis; PBL, peripheral blood lymphocytes; ShE, sheep erythrocyte; SACI, *Staphylococcus aureus* Cowan I bacteria; SDS, sodium dodecyl sulfate.

paque centrifugation, and depletion of monocytes by incubation in plastic culture plates (12). T cells were purified from PBL by nylon wool filtration (12). The T cell preparations contained less than 4% Ig^+ cells and 90% E rosette receptor-bearing cells. Enriched preparations of T cells bearing receptors for IgG Fc and IgM Fc were obtained after depletion of rosetted Tμ and Tγ cells, respectively, by Ficoll-Hypaque centrifugation (13).

Polyclonal xenoantisera and monoclonal xenoantibodies. Xenoantisera to framework determinants of HLA-A,B,C (14) or Ia-like antigens (15) and to β_2-μ (16) were prepared and characterized previously. The preparation and characterization of the monoclonal antibodies (Q5/13 and Q1/28) to framework determinants of Ia-like and HLA-A,B,C antigens, respectively, have been described elsewhere (17). Both antibodies are of the IgG2a subclass.

Rabbit antisera to ox erythrocyte (OxE) stroma were prepared as described (13). The IgM fraction of the antisera was isolated by Sephadex G-200 gel filtration, whereas the IgG fraction was isolated by DEAE cellulose chromatography. Xenoantisera to human and mouse Ig were elicited in rabbits by using conventional immunization procedures; anti-Ig antibodies were purified by affinity chromatography on Ig-Sepharose immunoadsorbents (18).

Rosette assays. Receptors for sheep erythrocytes (ShE) were detected by rosetting with 2-aminoethylisothiouronium bromide (AET) treated ShE (19). IgM and IgG Fc receptors were detected by rosetting with IgM antibody or IgG antibody-sensitized OxE, respectively (13). Membrane bound Ig (MbIg) and HLA antigens were detected by direct and indirect rosetting, respectively, by using purified anti-Ig antibody-coated E in an indirect rosette microassay (18). Simultaneous detection of cell receptors for ShE and Ia-like antigens on PHA-activated T cells was performed by rosetting T cells sensitized with antibodies to Ia-like antigens with an equal mixture of AET-ShE and chicken erythrocytes (ChE) coated with purified anti-mouse Ig antibodies. The percentage of rosettes binding both ShE and ChE was determined by microscopic examination of 300 cells.

Radioimmunometric assay. It was performed in microtiter plates as described previously (20) except for the addition of 1% bovine serum albumin in the medium used for washings of cells and the use of 5×10^5 cells per well.

Cell cultures and assay for DNA synthesis. PBL and T cells, 1×10^6/ml in RPMI 1640 medium containing 10% FCS, 0.1% PHA-P (Calbiochem, La Jolla, CA) and 1% Penn/strep were cultured for up to 96 hr at 37°C in an atmosphere of 5% CO_2. After mitogen stimulation, the cells were washed 3 times in MEM containing 0.5% α-methyl-D-mannoside in order to reduce cell-cell agglutination.

The mixed lymphocyte reaction (MLR) was performed essentially as described by Hartzman *et al.* (21). Monoclonal antibodies were added to the MLR at the initiation of culture. DNA synthesis was measured by pulsing triplicate cultures (0.2 ml) with 0.2 μCi ^{3}H-thymidine for 18 hr before harvesting on glass fiber filter paper with an automated sample harvester. The washed and dried filters were mixed with a liquid scintillation cocktail and then counted in a γ-scintillation counter.

Immunochemical analysis. Glycoproteins were purified by lentil-lectin affinity chromatography from nonidet P40 (NP40) extracts of ^{35}S-methionine intrinsically labeled PBL and T cells cultured with PHA for 72 hr (22). PBL and T cell glycoproteins reactive with xenoantibodies were immunoprecipitated with SACI, then eluted and electrophoresed in discontinuous 10% polyacrylamide slab gels containing 0.1% sodium dodecyl sulfate (SDS). The slab gel was prepared for fluorography as described by Bonner and Laskey (23).

RESULTS

Expression of Ia-like antigens on resting and PHA-activated T lymphocytes. Less than 4% of T cells either freshly isolated or cultured in the absence of PHA reacted with the MoAb Q5/13 to framework antigenic determinants of Ia-like antigens in the indirect rosette microassay (data not shown). Double labeling experiments whereby lymphocytes sensitized with the MoAb Q5/13 were rosetted with ChE coated with purified anti-human Ig antibodies and with ShE coated with purified anti-mouse Ig antibodies indicated that 98% of the Ia-like antigen-bearing cells in the unstimulated T cell preparations also expressed MbIg (data not shown). About 70% of T cells cultured with PHA for 72 hr reacted with the monoclonal antibody to Ia-like antigens; this reaction is specific for Ia-like antigens since it was blocked by pretreating PHA-stimulated T cells with a rabbit xenoantiserum directed to common structure(s) of Ia-like antigens but was not affected by pretreating the cells with a rabbit antiserum to common structure of HLA-A,B,C antigens. The Tμ and Tγ subpopulations treated with PHA were essentially equivalent in their ability to generate Ia-like antigen-bearing cells (Fig. 1).

Depletion of any Ia-like antigen bearing cells present in the nylon wool purified T cell preparation by first rosetting with MoAb Q5/13 followed by Ficoll-Hypaque centrifugation did not affect the subsequent PHA-induced expression of Ia-like antigens (data not shown), therefore indicating that the Ia-like antigen bearing T cells after PHA stimulation have not arisen from expansion of the small subpopulation of Ia-like antigen

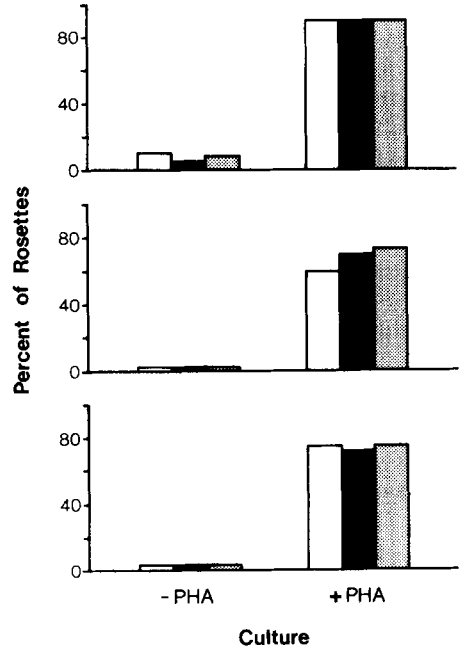

Figure 1. Analysis of human Tγ and Tμ lymphocyte subpopulations for PHA-induced expression of Ia-like antigens. Nylon wool purified T lymphocytes were separated into enriched preparations of Tγ and Tμ cells by depletion of IgM Fc receptor rosettes and IgG Fc receptor rosettes, respectively, on Ficoll-Hypaque medium. Purified T cells (□) and the Tγ (■) and Tμ (▨) subpopulation from 3 different donors (different panels) were cultured for 72 hr with and without PHA at 37°C and then tested for reactivity with monoclonal antibody Q5/13 to Ia-like antigens in an indirect rosette microassay.

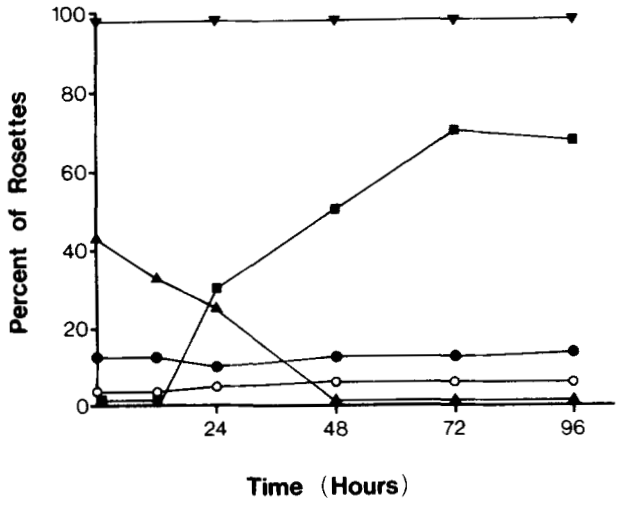

Figure 2. Effect of PHA activation on the expression of various human T lymphocyte surface markers. Nylon wool purified T lymphocytes cultured at 37°C for various times with PHA were tested for membrane bound Ig (○), Ia-like antigens (■), HLA-A,B antigens (▼), IgG Fc receptors (●) and IgM Fc receptors (▲) by various rosetting assays (see *Materials and Methods*).

bearing cells present in the unstimulated T cell preparations (<4%).

Kinetics of expression of Ia-like antigens and of other surface markers on PHA-activated T cells. The percentage of T cells reacting with MoAb Q5/13 increased as a function of time in culture with PHA, reaching a plateau after 72 hr (Fig. 2). The percentage of T cells expressing receptors for Fcγ remains unchanged throughout the 96 hr culture period with PHA as did the percentage of cells expressing MbIg and HLA-A,B,C antigens. In contrast, as the percentage of Ia-like antigen-bearing cells increased with time in culture with PHA, the percentage of cells expressing receptors for Fcμ decreased to zero by 48 hr of culture.

Sensitization of Ia-like antigen-bearing T lymphocytes with the MoAb Q5/13 and double rosetting with AET-ShE and with ChE coated with purified anti-mouse Ig xenoantibodies showed that the large majority of T cells express both Ia-like antigens and ShE receptors, and that treatment of T cells with anti-Ia-like antibodies did not block the detection of T cell receptors for ShE (Table I). Furthermore sensitizing Ia-like antigen-bearing T cells with the MoAb Q5/13 did not affect the detection of IgM Fc and IgG Fc receptors (data not shown).

Generation of the MLR with T cells stimulated by PHA. Although the T cells cultured in the absence of PHA exhibited few Ia-like antigen-bearing cells and were unable to stimulate in the MLR, T cells cultured from 12 to 96 hr with PHA showed increasing expression of Ia-like antigens correlating closely with increasing synthesis of DNA and increasing stimulatory activity in the MLR (Table II). The involvement of the newly expressed Ia-like antigen in the MLR, suggested by the parallel kinetics of Ia-like antigen expression and stimulatory activity in the MLR, was corroborated by MLR inhibition experiments with monoclonal antibodies to HLA antigens. Thus, MoAb Q5/13 to Ia-like antigens significantly inhibited the MLR performed with T cells or PBL as stimulators, whereas MoAb Q1/28 to HLA-A,B,C antigens or the P3 myeloma Ig had no effect on this reaction (Fig. 3). Similar results were observed when the MLR was performed in the presence of Fab_2 fragments from a xenoantiserum to Ia-like antigens (data not shown).

Structural analysis of Ia-like antigens on PHA-activated PBL and T cells. Immunoprecipitation of ^{35}S-methionine labeled glycoprotein from PBL and nylon wool purified T cells stimulated with PHA was performed with both polyclonal and monoclonal xenoantibodies to Ia-like antigens. Xenoantiserum No. 8612 immunoprecipitated four structures with the approximate m.w. of 100,000, 85,000, 34,000, and 29,000 from PHA-treated PBL (Fig. 4). The latter two components have the characteristic sizes of the α and β chains of the Ia-like antigens. The xenoantiserum No. 8612 immunoprecipitated three components of approximately 85,000, 34,000 and 29,000 m.w. from PHA-treated T cells. Evidence that the two smaller components detected in the T cell and PBL preparations were in fact the Ia-like α and β chain subunits was obtained when prior immunodepletion of the radiolabeled PBL or T cell glycoprotein preparation with a xenoantiserum to common determinants of Ia-like antigens (No. 8823) but not of HLA-A,B,C antigens (No. 9698) removed these two structures. The absence of the 100,000 m.w. structure from the T cell glycoprotein preparation suggests the exclusive synthesis of this molecule by B cells and/or macrophages and thus indicates the absence of these Ia-like

TABLE I

Simultaneous enumeration of PHA-stimulated human T cells for Ia-like antigens and sheep erythrocyte receptors

Donor	Double Rosetting Experiment[a]					
	A T cells[b]	B ShE alone	C ShE and ChE	D ChE alone	B + C / A	C / C + D
	%				%	
1	89	26	64	5	101	92
2	97	45	40	1	87	97
3	97	21	73	2	96	97
4	90	8	84	4	102	95

[a] Nylon wool purified T cells cultured with PHA for 72 hr were sensitized with MoAb Q5/13 to Ia-like antigens, washed, and then rosetted simultaneously with AET-sheep erythrocytes (ShE) and rabbit anti-mouse Ig-coated chicken erythrocytes (ChE).

[b] The percentage of T cells was determined by rosetting with AET-ShE.

[c] Defined as the percentage of T cells binding ShE after being sensitized with MoAb Q5/13 to Ia-like antigens in the double rosetting experiments.

[d] Defined as the percentage of Ia-like antigen-bearing cells that are T cells.

TABLE II

Kinetics of stimulatory capacity in allogeneic unidirectional MLR of T lymphocytes cultured in the presence and absence of PHA

Hr of Culture	Addition of PHA	DNA Synthesis	Ia-like Antigen[a]-Bearing Cells (% Rosettes)	Stimulation in MLR
		cpm		*cpm*
12	−	877	5	1,753
24	−	931	6	2,369
48	−	1127	9	2,151
72	−	1956	5	4,363
96	−	1911	2	3,987
12	+	1213	5	3,772
24	+	2,370	18	6,315
48	+	22,805	40	17,235
72	+	31,000	65	33,444
96	+	28,950	73	53,075

[a] Determined in an indirect rosette microassay with MoAb Q5/13.

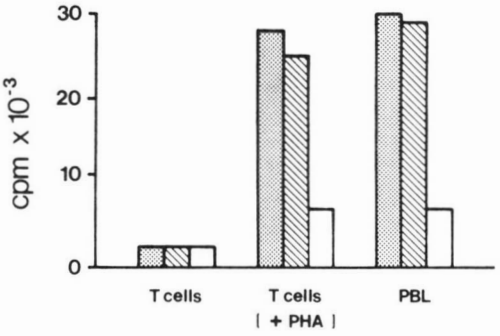

Figure 3. Human T lymphocytes as stimulator cells in the mixed lymphocyte reaction. Nylon wool purified T lymphocytes cultured with PHA for 72 hr as well as T cells and unseparated PBL (from the same individual) cultured for 72 hr in the absence of PHA were irradiated with 2000 rads and then used as stimulator cells in a MLR with allogeneic PBL. 1×10^5 stimulator cells and 1×10^5 responder cells were cultured in RPMI 1640 medium (200 μl) containing 50 μl of either monoclonal antibody Q5/13 to Ia-like antigens (□) HLA-A,B monoclonal antibody Q1/28 (▨), or the parent myeloma Ig (⊡) for 6 days in 96 well microtiter plates. 1 μCi of H-thymidine was added 18 hr before harvesting.

antigen bearing cells from our purified T cell preparations. It should be emphasized that the 85,000 m.w. component detected in the PHA-activated T cell preparation is not specific for T cells but can be found in some cultured B lymphoid cells as well as melanoma and carcinoma tumor cell lines (data not shown). The Victor B lymphoid cell line synthesizes Ia-like antigens but little if any of the 100,000 and 85,000 m.w. components detected by xenoantiserum No. 8612; this cell line was therefore used as a reference for the mobility of Ia-like antigens in SDS-PAGE (Fig. 4).

Interestingly, the amounts of Ia-like antigens immunoprecipitated from the PHA-activated T cells is considerably less than that detected for the PHA-treated PBL. A similar result was observed at the cell surface where PHA-activated T cells bound about 5 times less anti-Ia-like monoclonal antibody (No. Q5/13) than did PHA-activated PBL, as assessed in a ^{125}I-radioimmunometric binding assay (data not shown). It should be noted that immunoprecipitation of PHA activated T cells glycoprotein preparations with the MoAb to Ia-like antigens Q5/13 detected only the 34,000 and 29,000 m.w. components of Ia-like antigens (Fig. 5). The light component displayed the characteristic change in mobility under nonreducing conditions, previously described for the β chain.

DISCUSSION

The percentage of Ia-like antigen-bearing cells in our unstimulated T cell preparations was from 0 to 5% in agreement with other investigators (1, 5, 9, 10). However, virtually all (98%) of these Ia-like antigen bearing cells expressed membrane Ig and were therefore of B cell rather than T cell origin. The presence of a small subpopulation of Ig^+ B cells that fail to bind nylon wool and thus contaminate nylon wool T cell preparations has been previously reported (14). Although we cannot exclude that there is a small subpopulation of unstimulated T cells expressing Ia-like antigens, this subpopulation must be less than 1% of our purified T cell preparation. The percentage of our purified T cells rosetting with antibodies to Ia-like antigens is lower than that detected by other investigators who used similar reagents and serologic approaches but isolated T peripheral lymphocytes by rosetting peripheral lymphocytes with neuraminidase-treated ShEs and differential centrifugation on Ficoll-Hypaque (3–5).

In contrast to the results with unstimulated T cells, Ia-like antigens were easily detected on the majority of PHA-activated T cells (~70%). This expression correlated closely with an increase in DNA synthesis that is in contrast to the delayed expression of Ia-like antigens reported by Ko *et al.* (6). The earlier detection of Ia-like antigens in our experiments may be due to our use of a highly sensitive rosette assay rather than the immunofluoresence assay used by Ko et al. (6).

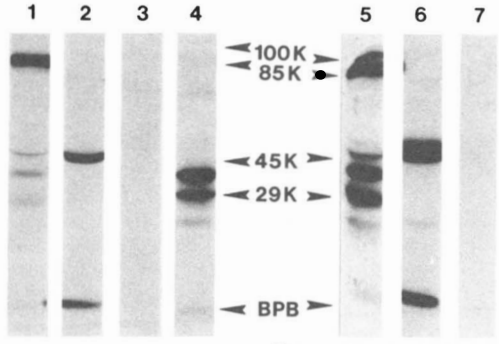

Figure 4. SDS-PAGE analysis of ^{35}S-methionine-labeled glycoproteins immunoprecipitated from PBL and PHA-activated lymphocytes with HLA xenoantisera. ^{35}S-methionine-labeled glycoproteins were isolated from intrinsically labeled PHA-activated T cells and unfractionated PBL first by solubilization in nonionic detergent (NP_{40}) followed by purification on lentil-lectin Sepharose immunoadsorbents. ^{35}S-glycoproteins from PHA-activated T cells (lanes 1–3) and PBL (lanes 5–7) were immunoprecipitated with polyclonal xenoantiserum No. 8612 to Ia-like antigens (lanes 1 and 5), anti-β_2-μ xenoantiserum (lanes 2 and 6) and normal rabbit serum (lanes 3 and 7). Lane 4 shows ^{2}H-phenylalanine-labeled Ia-like antigens immunoprecipitated from the Victor B lymphoid cell glycoprotein with xenoantiserum No. 8612. The immunoprecipitates were adsorbed on to SACI, washed, eluted, and electrophoresed on a SDS-polyacrylamide slab gel.

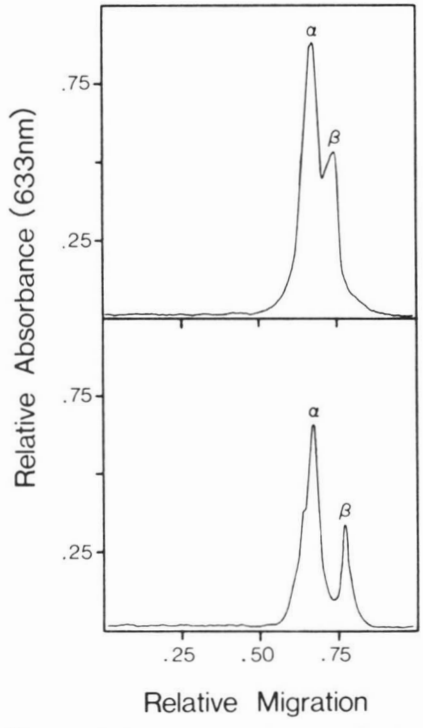

Figure 5. M.w. analysis of Ia-like antigens synthesized by human T lymphocytes. ^{35}S-methionine-labeled glycoproteins isolated from nylon wool purified T cells activated by PHA were reacted with monoclonal antibody Q5/13 to Ia-like antigens. The immunoprecipitates were electrophoresed under reducing (*upper graph*) and nonreducing conditions (*lower graph*) by SDS-PAGE. The fluorograph was scanned at 633 nm.

The expression of Ia-like antigens on PHA-activated T cells occurs by induction rather than by clonal expansion or by binding of shed B cell Ia-like antigens since depletion of any Ia-like antigen bearing cells from T cell preparation had no effect on the subsequent PHA-induced expression of Ia-like antigens. These data are consistent with a recent study by Reinherz *et al.* (8) in which cytotoxicity was used to deplete Ia-like antigens bearing cells from a T cell preparation.

Sensitization of PHA-activated T cells with monoclonal antibodies to Ia-like antigens had no effect on the detection of sheep erythrocyte receptors and of IgFcγ and IgFcμ receptors, suggesting a lack of spatial association between Ia-like antigens and IgFc receptors on T cells (1, 5). It was interesting to note that the appearance of Ia-like antigens on PHA-activated T cells coincided with the disappearance of receptors for IgM Fc. While this work was in progress, Lydard and Fanger (24) also reported the disappearance of IgM Fc receptors from mitogen-treated T cells. The reason for this loss of receptors is at present unclear, however, prior expression of IgM Fc receptors is not a requirement for subsequent Ia-like antigen expression since both the IgM Fc and the IgG Fc receptor subpopulations of T cells will express Ia-like antigens when cultured with PHA.

PHA-treated T cell preparations were found to stimulate allogeneic T cells in a unidirectional MLR. The possibility that the T cell MLR was effected by presentation of PHA bound to the stimulated T cells is unlikely since 1) the stimulator cells were washed extensively with α-methyl-D-mannoside to remove lectin before the MLR and 2) optimal binding of PHA to lymphocytes at 37°C occurs within 30 min after its addition (25), whereas the stimulator T cells required at least 24 hr in culture with PHA to effect the MLR. Evidence for the involvement of Ia-like antigens in this reaction was that the degree of MLR stimulation correlated closely with the percentage of Ia-like antigen bearing T cells and the reaction was specifically blocked by monoclonal antibodies to Ia-like antigens. The possibility that this blocking may have occurred through an antibody-dependent cell-mediated cytotoxic event (26) is unlikely since 1) monoclonal antibodies to HLA-A,B,C antigens, which were of the same IgG subclass as the antibodies to Ia-like antigens, failed to block the MLR and 2) the blocking was also achieved with Fab_2 fragments from a polyclonal xenoantiserum to Ia-like antigens. These results suggest that one function of Ia-like antigens, involvement in the MLR, is similar for Ia-like antigens of B and T cells. This conclusion is also supported by the ability of mitogen-activated Ia-like antigen bearing T cell to stimulate autologous T cells in a MLR (27).

Indirect immunoprecipitation of intrinsically labeled glycoproteins from PHA-activated T cells indicated that they synthesize Ia-like antigens that have the apparent m.w. of B cell-derived Ia-like antigens. The lighter component of the T cell-derived Ia-like antigens exhibits an increased mobility when electrophoresed under nonreducing conditions like that reported for the β chain of B cell-derived Ia-like antigens (for review see 28). The low level of Ia-like antigens detected in the PHA-treated T cell glycoprotein preparations raises the possibility that Ia-like antigens are not synthesized by PHA-activated T cells but are passively adsorbed from the culture medium or have a different structure than that of B cell-derived Ia-like antigens (29). The former possibility is unlikely since our experiments were performed with purified preparations of T cells whose purity was verified by cell-surface marker analysis and by immunochemical criteria and since experiments designed to measure T cell uptake of Ia-like antigens in culture fluids have given negative results (6, 7). The different structure hypothesis is also unlikely since the low level of Ia-like antigens detected immunochemically in PHA-activated T cells is consistent with a low level of Ia-like antigens expressed on the surface of these cells, and two different monoclonal antibodies to Ia-like antigens immunoprecipitated only 34,000 and 29,000 m.w. components from PHA-activated T cells. In summary, these data indicate that T cells activated by PHA synthesize detectable levels of Ia-like antigens that are structurally and functionally similar to B cell-derived Ia-like antigens and suggest an importance of these antigens to T cell-activated functions.

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