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Antigen presentation by common variable immunodeficiency (CVID) B cells and monocytes is unimpaired

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SUMMARY

CVID is a primary immunodeficiency syndrome comprising a heterogeneous group of patients with hypogammaglobulinaemia and defective formation of specific antibodies. Previous studies demonstrated defective T cell responsiveness to antigen in a major subgroup of patients. In the present study we investigated the capacity of peripheral blood monocytes and Epstein–Barr virus (EBV)-transformed B cell lines from seven patients with CVID, including two patients expressing an extended MHC haplotype described to be associated with CVID, to present antigen (Tet. Tox.) to $CD4^+$ antigen-specific T cell lines from healthy controls. The results presented show an unimpaired capacity of peripheral blood monocytes to present antigen in all patients studied. In addition, the present study demonstrates for the first time that CVID B cells function normally as antigen-presenting cells (APC). These findings indicate that expression of a certain MHC phenotype in CVID is not associated with a defect in the presentation of recall antigen by monocytes and B cells. Based on these studies, uptake, processing and re-expression of recall antigen in association with MHC class II molecules on the APC surface are functional and there is no indication for structural abnormalities of the MHC class II molecules expressed by the patients studied that could be essential for their function in antigen binding and presentation.

Keywords common variable immunodeficiency antigen presentation B cells monocytes

INTRODUCTION

The first step in T cell activation is recognition of antigen on the surface of antigen-presenting cells (APC) in association with products of the MHC gene complex. Professional APC, such as dendritic cells, cells of the mononuclear phagocyte system and B lymphocytes, convert ingested protein antigens to peptides (antigen processing) and present peptide–MHC complexes in a form that can be recognized by T lymphocytes [1–3]. Surface membrane structures of the APC and APC-derived cytokines provide costimulatory activity to the T cell [2–4], thereby leading to fully functional T cell activation which enables T cells to promote B cell activation, proliferation, differentiation and immunoglobulin production [5,6].

CVID is a primary immunodeficiency syndrome characterized by hypogammaglobulinaemia and defective formation of specific antibodies. Evidence available at present indicates that in most of these patients the B cells are principally capable of making antibodies and suggests that the defect in antibody production is not, in the great majority of cases, at the B cell level. Several abnormalities at the T cell level have been described in CVID patients, and recent evidence suggests that a partial defect in T cell activation is present in most patients with CVID, thereby leading to an impairment in T–B cell interaction required for normal antibody formation. A defective response of T cells to recall antigen has been described in a major subset of CVID patients [7,8]. Impaired responsiveness of T cells to stimulation with recall antigen could be due to a defect at the level of the $CD4^+$ T cell, the major subset responsible for recall antigen responses, such as impaired signal transduction associated with T cell receptor (TCR)-mediated T cell activation [9], or it could be explained by a defect at the level of the APC, such as inadequate uptake and/or processing of exogenous antigen by the APC or functional abnormalities of MHC class II molecules resulting in weak or unstable binding of antigenic peptides to MHC class II, leading to impaired activation of antigen-specific T cells of the $CD4^+$ phenotype.

Susceptibility to certain immune-mediated diseases such as insulin-dependent type I diabetes mellitus (IDDM), ankylosing spondylitis or progression of disease in HIV-1 infection has been

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related to the expression of specific amino acids or epitopes within the HLA-DR, -DP or -DQ molecules. In IDDM patients the increased frequency of certain MHC class II genes may have a functional role in the development of disease [10–12], as in the mouse the development of T cell-mediated autoimmune diabetes is closely linked to the expression of a certain MHC class II phenotype [13,14]. A functional correlate was provided by the demonstration of weak and unstable binding of immunogenic peptides to the MHC class II molecule, resulting in impaired antigen presentation, ultimately leading to failure in the elimination and/or inactivation of autoreactive T cells [15]. Alternatively, a genetic contribution of MHC class II to IDDM in the BB rat model has been suggested, i.e. that binding of β cell peptides (the autoantigen) to certain permissive MHC class II molecules is likely to result in the activation of diabetes-inducing T cells [16]. Thus a functional correlate for disease association with certain MHC alleles may arise on the basis of abnormalities in antigen presentation by certain MHC class II phenotypes.

Previous studies showed an association of CVID with certain alleles encoded in the MHC region [17–23]. Analysis of extended MHC haplotypes (e.g. HLA-DQB1*0201, -DR3, C4B-Sf, C4A-0, G11-15, Bf-04, C2a, HSP-7.5, TNFα-5, HLA-B8, -A1) suggests that CVID may be associated with genes of the MHC class II region such as HLA-DQβ [17] or the class III region [19,20]. An association of CVID with certain MHC haplotypes could point towards a possible defect at the level of the function of MHC on the APC. Results of studies on antigen presentation by APC of CVID patients are not uniform, and this could be due to the heterogeneity of the CVID population with respect to the expression of a certain MHC phenotype [24,25]. Therefore, antigen presentation in this group of patients needs further clarification.

The aim of the present study was to clarify further whether a defect in antigen presentation is present in CVID, and to answer the question whether a possible defect would be associated with the expression of certain MHC class II phenotypes. We studied seven patients with different MHC phenotypes, including two patients expressing an extended MHC haplotype, and examined antigen presentation restricted by different HLA-DR haplotypes. The results presented show that CVID monocytes present recall antigen normally, independent of the MHC class II phenotype expressed by the patients. Furthermore, we have proved for the first time that also B cells from CVID patients present antigen normally.

PATIENTS AND METHODS

Patients

Seven patients with well documented CVID according to the diagnostic criteria of the WHO expert group for primary immunodeficiency diseases were included in the study [26]. Patients were on regular substitution therapy with IVIG at 3- to 4-week intervals. Blood samples were always collected before IVIG infusion was given. All patients were immunized and re-boosted with tetanus vaccine before the study was started.

Cell preparation

Mononuclear cells (MNC) were isolated from heparinized peripheral blood (7·5 U/ml preservative-free heparin) by buoyant density gradient centrifugation (Lymphoprep; Nycomed Pharma AS, Oslo, Norway). Monocytes were prepared by adherence to plastic surfaces as described earlier [27]. Cell purity was > 85% as determined by measurement of $CD14^+$ cells with flow cytometry. Non-adherent cells were fractionated into T- and B-enriched cells by rosetting with 2-aminoethylisothiouronium bromide (Sigma Chemical Co., St Louis, MO) as described earlier [27].

Cell lines

B cells contained in the non-T cell population were transformed with Epstein–Barr virus (EBV) using supernatant from a B 95-8 marmoset cell line (ATCC, Rockville, MD) according to a standard protocol [28]. Growing cells were expanded in complete RPMI 1640 medium [27] containing 20% heat-inactivated fetal calf serum (FCS; HyClone Labs, Logan, UT) under standard conditions.

Antigen-specific T cell lines were established from healthy Tet. Tox.-immunized blood donors sharing one HLA-DR haplotype with one or more CVID patients. MNC were stimulated in complete RPMI 1640 medium with tetanus toxoid (10 Lf/ml; Swiss Serum and Vaccine Institute, Bern, Switzerland) for 7 days. Cells were then further cultured in the presence of highly purified IL-2 (10 U/ml; Lymphocult-T-HP; Biotest AG, Dreieich, Germany). After at least 14 days of culture the cells were repeatedly restimulated with freshly isolated γ-irradiated autologous macrophages in the presence of tetanus toxoid and IL-2. Antigen-specific T cells were further cultured by limiting dilution (U-bottomed plate; Greiner, Gmünd, Austria) in the presence of tetanus toxoid, irradiated autologous MNC and IL-2 (10 U/ml). One week later, half of the medium was replaced by fresh IL-2-containing medium. Growing cells were transferred to 24-well plates and expanded. Antigen specificity was tested by stimulating the T cell lines with *Escherichia coli* (5×10^6/ml, heat-inactivated *E. coli* 089:H10), purified protein derivative (PPD) (5 μg/ml; Tuberculin PPD; Statens Seruminstitut, Copenhagen, Denmark), and tetanus toxoid (10 Lf/ml; Swiss Serum and Vaccine Institute), and the absence of alloreactivity was examined by stimulation with HLA-mismatched APC from healthy blood donors. The phenotype of the cell lines was tested by flow cytometry using commercially available MoAbs and standard methodology.

Antigen-specific T cell lines were used as responder cells in the experiments with APC from patients at least 14 days after the last stimulation with irradiated autologous APC and antigen.

T cell proliferation and cytokine production

Triplicate cultures containing monocytes (γ-irradiated with 60 Gy, 1×10^4/well) or EBV-transformed B cells (γ-irradiated with 90 Gy, 5×10^4/well) and peripheral blood T-enriched cells (1×10^5/well, purity > 90% $CD3^+$ cells) or antigen-specific T cell lines (5×10^4/well) were resuspended in complete RPMI 1640 medium containing 10% pooled, heat-inactivated human AB serum and were cultured in flat-bottomed microtitre plates (0·2 ml/well; Falcon microtest II; Becton Dickinson, Lincoln Park, NJ). Tetanus toxoid (10 Lf/ml) or phytohaemagglutinin (PHA; 1:1250; Wellcome, Dartford, UK) were added and cells were cultured for 3 days (antigen stimulation of T cell lines and PHA stimulation of resting peripheral blood T cells) or for 7 days (antigen stimulation of resting T cells) at 37°C in a CO_2 incubator (5% CO_2 in humidified air). T cell proliferation was assessed by measuring ^{3}H-thymidine incorporation as described earlier [8]. IL-2 release was measured with the Interleukin two Enzyme Immunoassay Kit (Immunotech S.A., Marseilles, France), and interferon-gamma (IFN-γ) with the IFN-gamma EASIA Kit (Medgenix Diagnostics, Fleurus, Belgium) as described earlier [8].

Table 1. Characterization of the patients

Patient	Serum immunoglobulin (g/l)			Lymphocyte phentotypes (%)				^{3}H-thymidine incorporation (dpm×10^3)			HLA-				
	IgG	IgA	IgM	CD3	CD4	CD8	CD19	Tet. Tox.	PHA	Medium	A	B	C	DR	DQ
1	0·63	<0·01	<0·01	82	26	61	7	1·2	128	0·7	1	8	w7	3	2
2	0·05	<0·01	0·13	78	24	58	7	1·6	199	0·2	1	8	w7	3	2
3	2·17	0·20	0·01	74	41	41	11	0·9	78	0·2	1, 2	13, w47	w6	4, 7	2, 3
4	1·72	0·34	0·11	72	31	44	11	1·8	93	0·6	2, 19(30)	13, 27	w2, w6	4, 7	2, 3
5	1·87	<0·01	0·27	70	36	28	16	2·1	96	0·5	2, 9(24)	16(38), 35	w4	4, 7	ND
6	0·83	<0·01	0·57	80	23	59	11	1·7	128	0·2	11	5(51), 40(60)	w3	4	3
7	0·75	<0·01	0·24	76	36	39	5	1·3	81	0·2	2	12	w5	2, 5	ND
Patients ($n = 7$)	1·1*	0·1*	0·2*	76	31**	47***	10	1·5*	115	0·4					
Mean ± s.d.	± 0·8	± 0·1	± 0·2	± 4	± 7	± 12	± 4	± 0·4	± 42	± 0·2					
Controls ($n = 7$)	11·4	2·4	1·5	70	44	31	12	22	122	0·7					
Mean ± s.d.	± 1·9	± 0·6	± 0·5	± 7	± 5	± 7	± 6	± 6	± 33	± 0·6					

Serum immunoglobulin levels were determined before IgG replacement therapy was started. Antigen- or mitogen-induced T cell proliferation was examined by measuring ^{3}H-thymidine incorporation following stimulation of T-enriched cells (1×10^5) with tetanus toxoid (10 Lf/ml) or phytohaemagglutinin (PHA; 1:1250) in the presence of autologous monocytes (1×10^4) as described in Patients and Methods. Statistically significant differences: *$P < 0{\cdot}001$; **$P = 0{\cdot}002$; ***$P = 0{\cdot}012$ (Student's *t*-test).

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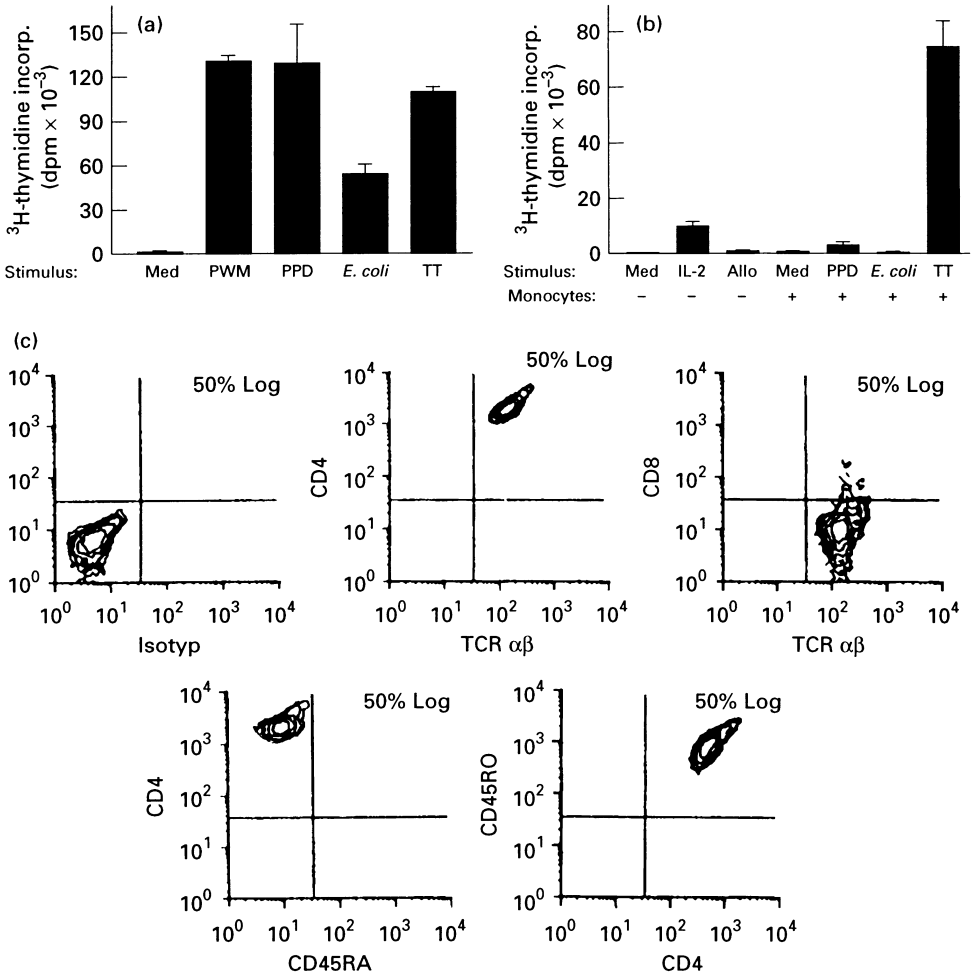

Fig. 1. Characterization of an HLA-DR4-restricted Tet. Tox.-specific T cell line from a healthy control. The Tet. Tox.-specific T cell line was established from the peripheral blood mononuclear cell (MNC) population of a Tet. Tox.-immunized healthy donor as described in Patients and Methods. The proliferative response of peripheral blood MNC (a) or the antigen-specific T cell line (b) to stimulation with different recall antigens, alloantigen or mitogen (pokeweed mitogen (PWM)) was examined as described in Patients and Methods. The phenotype of the T cell line was examined by two-colour flow cytometry (c). PPD, Purified protein derivative.

FACS analysis

The purity and lineage of cell preparations were tested in two-colour flow cytometry analysis using a FACScan (Becton Dickinson, San Jose, CA). Anti-TCR $\alpha\beta$ (TCR-α/β-1), anti-CD3 (Leu-4), anti-CD4 (Leu-3a), anti-CD8 (Leu-2a), anti-CD45RA (Leu-18), anti-CD45RO (Leu-45RO), anti-CD19 (Leu-12), anti-CD20 (Leu-16), and anti-CD14 (Leu-M3), all MoAbs purchased from Becton Dickinson, were used in direct immunoflorescence staining following a standard methodology.

Statistical analysis

Statistical analysis was performed using the two-tailed Student's *t*-test for unpaired samples, and $P < 0{\cdot}05$ was considered significant.

RESULTS

A defective T cell response to antigen in CVID patients is not limited to the expression of a certain MHC class II phenotype

As can be seen in Table 1, the seven patients included in the study had the typical characteristics of CVID, i.e. significant reduction of one or more serum immunoglobulin isotypes, normal numbers of peripheral blood T and B lymphocytes, and a variable increase in $CD8^+$ cells with a normal or slightly reduced percentage of $CD4^+$ cells. In all seven patients studied there was a significant defect in their T cells' capacity to respond to antigen stimulation (tetanus toxoid) despite recent vaccination before the study, while PHA responses were normal, as has also been described previously [8,29,30]. The seven patients showed four different HLA-DR characteristics. Defective T cell response was observed in patients independently of their MHC class II phenotype, including two unrelated patients (patients 1 and 2) who were HLA-histoidentical and expressed an extended MHC haplotype (Table 1).

Independent of the patients' MHC class II phenotype expressed, the capacity of patients' peripheral blood monocytes to function as APC is normal

To clarify whether patients' defective T cell response to antigen was due to a defect at the level of the APC, we performed experiments using antigen-specific T cell lines from healthy blood donors as responder cells and, as APC, adherence-purified peripheral blood monocytes from unrelated healthy controls or CVID patients sharing at least one HLA-DR haplotype with the T

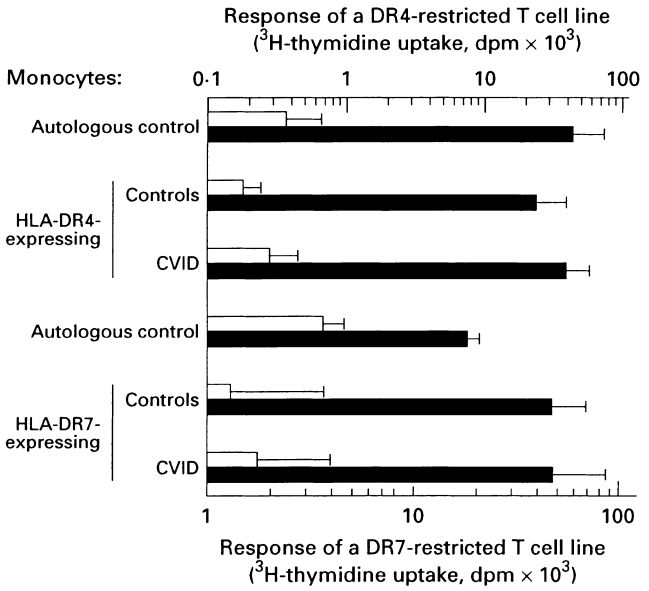

Fig. 2. Response of an HLA-DR4- or DR7-restricted normal T cell line to Tet. Tox. presented by monocytes from patients and controls. HLA-DR4- or DR7-expressing monocytes (1×10^4/well, γ-irradiated) and Tet. Tox.-specific HLA-DR4- or DR7-restricted antigen-specific T cells (5×10^4/well) were stimulated with Tet. Tox. (10 Lf/ml) for 3 days before ^{3}H-thymidine incorporation was determined. Monocytes from four patients expressing HLA-DR4 (top), three patients expressing HLA-DR7 (bottom), two HLA-DR4-expressing and two HLA-DR7-expressing haploidentical controls were tested. Values represent mean ± s.d. of six to seven experiments (patients) or three to four experiments (haploidentical controls). As a control, autologous monocytes were used to present antigen to the respective DR-7- or DR-4-expressing T cell line (mean ± s.d. of six experiments for DR4-expressing T cell line, mean ± s.d. of one representative experiment set up in triplicate for the HLA-DR7-expressing T cell line). □, Medium; ■, Tet. Tox.

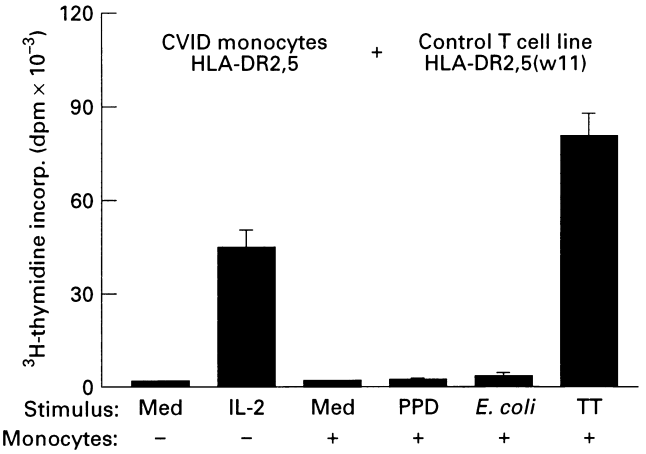

Fig. 3. Antigen presentation of HLA-DR2,5-expressing CVID monocytes to a DR2-restricted tetanus toxoid-specific normal T cell line. Monocytes (1×10^4/well, γ-irradiated) from a CVID patient expressing HLA-DR2,5 and Tet. Tox. antigen-specific HLA-DR2-restricted normal T cells (5×10^4/well) were stimulated with Tet. Tox. (10 Lf/ml), *Escherichia coli* (5×10^6/ml), purified protein derivative (PPD; 5 μg/ml) or IL-2 (10 U/ml). ^{3}H-thymidine incorporation was determined after 3 days. Values represent mean ± s.d. of one representative experiment set up in triplicate.

cell donor. Figure 1 shows the characteristics of a representative antigen-specific HLA-DR4-restricted T cell line from a healthy person. The cell line was specific for Tet. Tox., and in contrast to the donor's peripheral blood T cells (Fig. 1a) showed practically no response to stimulation with alloantigen or other recall antigens such as *E. coli* or PPD in the presence of autologous monocytes (Fig. 1b). The phenotype of the antigen-specific T cell lines generated was uniformly $CD4^+$, $CD3^+$, TCR $\alpha\beta^+$, $CD45RO^+$, $CD45RA^-$, $CD8^-$ (for a representative example see Fig. 1c).

The results shown in Fig. 2 indicate that adherence-purified peripheral blood monocytes from healthy donors were capable of presenting antigen to a Tet. Tox.-specific T cell line sharing one HLA-DR haplotype (e.g. DR4-positive; DR7-positive). The proliferative response obtained upon stimulation with haploidentical monocytes in this system was comparable to the response observed when autologous monocytes were added as APC. In addition, our results clearly show that monocytes from CVID patients functioned normally with respect to presentation of antigen in this system (Fig. 2). CVID monocytes that expressed either DR4 or DR7 were capable of presenting antigen to Tet. Tox.-specific DR4- or DR7-expressing T cells, and the proliferative response obtained was comparable to that observed with normal HLA-DR-compatible monocytes as APC. Allo-non-responsiveness of the antigen-specific T cell lines has been reconfirmed in every experiment by showing that the proliferative response of T cells cocultured with APC from patients or healthy controls sharing one HLA-DR haplotype with the T cells never exceeded T cell proliferation observed in cultures containing autologous instead of HLA-DR-haploidentical APC (Fig. 2). Furthermore, monocyte

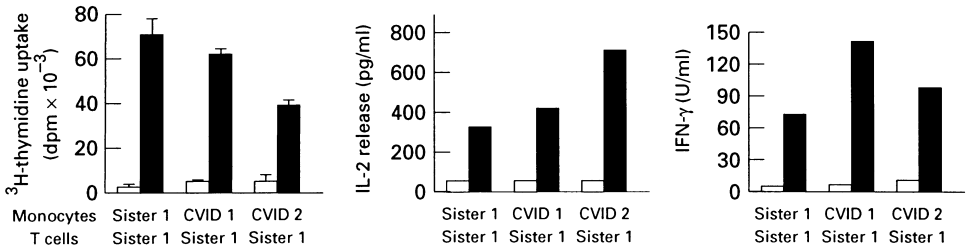

Fig. 4. Monocytes from two CVID patients expressing an extended MHC haplotype present antigen normally to histoidentical T cells. Monocytes from two unrelated CVID patients expressing an extended MHC haplotype and Tet. Tox.-specific T cells from the histoidentical healthy sister of patient 1 were cocultured in the presence of Tet. Tox. (10 Lf/ml) as described in Patients and Methods. IL-2 and IFN-γ release were measured in the cell-free supernatants by ELISA after 48 h of incubation, and the proliferative response was determined by measuring ^{3}H-thymidine incorporation after 3 days. Values shown are from one representative experiment (proliferative response is given as mean ± s.d. of triplicate cultures). □, Medium; ■, Tet. Tox.

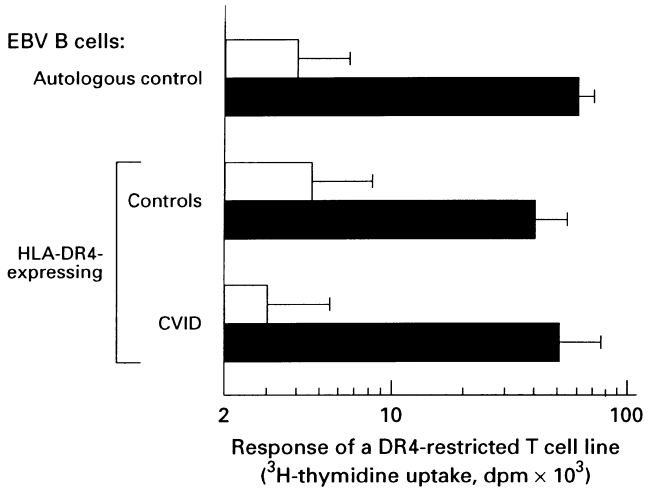

Fig. 5. CVID patients' B cells present antigen normally. A Tet. Tox.-specific HLA-DR4-restricted T cell line was stimulated with antigen (Tet. Tox. 10 Lf/ml) in the presence of Epstein–Barr virus (EBV)-transformed B cells (5×10^4/well, γ-irradiated). Four patients expressing HLA-DR4 and three healthy haploidentical controls expressing DR4 were examined. Proliferative responses were determined by measuring ^{3}H-thymidine uptake, and values represent mean ± s.d. of nine (patients) and three (controls) experiments. As a control, autologous EBV-transformed B cells were used to present antigen to the T cell line, and the results given represent the mean ± s.d. of five experiments. □, Medium; ■, Tet. Tox.

antigen presentation was also normal in a patient expressing DR2 when antigen-specific stimulation of a DR2-restricted normal T cell line was examined (Fig. 3). No response was observed when the T cell lines were cultured in the presence of monocytes alone, when stimulated with antigen in the absence of APC, or following addition of irrelevant antigens such as PPD or *E. coli*.

The results depicted in Fig. 4 extend the findings presented above by showing that monocytes from two additional patients expressing an extended MHC haplotype described to be associated with CVID presented Tet. Tox. normally to a histoidentical antigen-specific T cell line established from the healthy sister of patient 1. The proliferative response as well as IL-2 and IFN-γ release following stimulation of the T cells with antigen in the presence of the patients' monocytes was comparable to the levels observed when autologous monocytes were employed as APC.

These results clearly show that monocytes from CVID patients function normally with respect to presentation of antigen to T cells from healthy individuals, and that the capacity to present recall antigen was independent of the HLA-class II haplotype expressed and acting as the restriction element.

B cells from patients with CVID function normally as APC

In addition to peripheral blood monocytes we examined the capacity of EBV-transformed B cells from patients to present antigen to a Tet. Tox.-specific T cell line obtained from a healthy control. The results presented in Fig. 5 show that both autologous and HLA-DR-compatible EBV-transformed B cells from patients or control individuals expressing HLA-DR4 presented antigen to the HLA-DR4-positive T cell line, and the levels of proliferative response obtained were comparable between patients and controls. Allo-non-responsiveness of the antigen-specific T cell lines was again reconfirmed by showing that the proliferative response of T cells cocultured with APC from patients or healthy controls sharing one HLA-DR haplotype with the T cells never exceeded T cell proliferation observed in cultures containing autologous cells only (Fig. 5). Furthermore, EBV-transformed B cells from the two patients with the extended haplotype were examined for their antigen-presenting capacity. Normal levels of proliferative responses were observed when a Tet. Tox.-specific T cell line from the healthy histoidentical sister of one patient was stimulated with antigen in the presence of the patients' EBV-transformed B cells (antigen-induced ^{3}H-thymidine incorporation in the normal T cell line stimulated in the presence of EBV-B cells from: patient 1, 36 291 ± 3203 dpm (mean ± s.d. of triplicate cultures); patient 2, 33 358 ± 8835 dpm; HLA-DR compatible healthy control, 17 314 ± 1441 dpm).

DISCUSSION

Antigen processing and presentation is an important early event in the course of the immune response. Only a few studies have formally examined the capacity of CVID APC to present recall antigen, and the results from these studies are inconclusive, as defective antigen presentation has been observed under some circumstances, but not in others [24,25,31–33]. In the present study we further clarified the functional integrity of APC (both monocytes and B lymphocytes) in CVID patients belonging to a major subgroup in whom antigen-induced T cell activation was shown to be defective. We established Tet. Tox.-specific $CD4^+$ T cell lines generated from MHC class II-compatible healthy controls as responder cells for MHC class II-dependent antigen presentation to study the function of APC of CVID patients independent of a possible defect in antigen responsiveness at the level of the T cell. In addition, this system enabled us to use the same T cell line as responding cells for antigen presented by patients and controls as well as monocytes and B cells, thus allowing a direct comparison between patients and controls as well as the different types of APC. Our results show that the capacity of peripheral blood monocytes and EBV-transformed B cells from CVID patients to present antigen in conjunction with MHC class II to $CD4^+$ T cells was unimpaired in all seven patients studied. Thus a defect in uptake, processing and re-expression of the antigen in association with MHC class II molecules on the surface of the APC as well as structural abnormalities of the MHC class II molecules relevant for their function in antigen binding and presentation could be ruled out. This conclusion is in accordance with recent findings from our laboratory showing a defect in signal integration in these patients' purified $CD4^+$ or $CD8^+$ T cells when stimulated with MoAbs against defined surface structures in the absence of accessory cells (V. Thon, manuscript submitted).

CVID and IgA deficiency are the most prevalent immunodeficiencies and usually occur sporadically. However, reports of familial aggregation of cases of CVID and IgA deficiency continue to accumulate [18–20,22,23,34] and have led to the notion that these patients might share genetic factors influencing disease susceptibility, including MHC genes; however, this correlation has not been established without exception [19,20,22]. Although polymorphic MHC genes have a profound influence on various levels of the immune system, the mechanism by which the expression of certain MHC haplotypes might correlate with a defect in antibody production is unclear. The results presented in this study clearly indicate that independent of the MHC phenotype expressed, patients' monocytes show a normal capacity to present

recall antigen in association with MHC class II to $CD4^+$ antigen-specific T cells from an HLA-DR-compatible healthy donor. In addition, our findings indicate that expression of the extended MHC haplotype in CVID is not associated with functional abnormalities of the MHC class II molecules with respect to presentation of a recall antigen such as tetanus toxoid.

Reduced levels of immunoglobulin in CVID have been attributed to a failure of B cells to mature into immunoglobulin-secreting cells due to the lack of appropriate T cell help or an intrinsic B cell defect [35]. In addition to antibody production, B lymphocytes play an important role in the immune system by acting as APC, and cell-specific regulatory mechanisms have been shown to have an impact on antigen presentation [27]. Although the presence of intrinsic B cell defects in CVID patients has been discussed over the years and a variety of functional abnormalities in the B cells of these patients have been described [35–37], the capacity of CVID B cells to function as APC has not been investigated previously. The present study extends the functional characterization of B cells in CVID by showing that EBV-transformed B cells from CVID patients function normally as APC for MHC class II-dependent $CD4^+$ T cell activation. These findings provide further evidence that important B cell functions operational in T–B cell interaction required for a normal antibody response, such as MHC class II-dependent presentation of antigen to $CD4^+$ T cells [1], are principally intact in CVID patients with an impaired T cell response to antigen. More likely, the defective antibody production present in these patients could be explained by a defect at the T cell level, such as a partial T cell defect associated with TCR-dependent T cell activation, leading to impaired help for B cell antibody production [5,6,8].

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