Clin. exp. Immunol. (1983) **51**, 600–604.

IgG2 deficiency in a healthy blood donor. Concomitant lack of IgG2, IgA and IgE immunoglobulins and specific anti-carbohydrate antibodies

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(Accepted for publication 1 October 1982)

SUMMARY

Lack of serum IgG2, IgA and IgE was found in a healthy male adult blood donor. No secretory IgA could be demonstrated. *In vitro* activation of lymphocytes did not induce IgA secreting cells although no class specific suppressor cells could be found. Normal or slightly subnormal titres to a variety of bacterial and viral antigens were demonstrated whereas anti-carbohydrate antibodies (anti-teichoic acid, anti-dextran and anti-pneumococcal polysaccharide) were virtually absent. Isoagglutinins and heteroagglutinins were present in somewhat lower concentrations than normal.

INTRODUCTION

Since the description of IgG subclasses, a variety of immunodeficiency disorders have been described where selective IgG subclass deficiency has been accompanied by a substantially raised incidence of bacterial infections (for review see Oxelius, 1979b). Although lack of IgG1, IgG3 and IgG4 may be seen in healthy individuals (van Loghem *et al.*, 1980), selective IgG2 deficiency results in a markedly increased susceptibility to infections. Therefore, it has been suggested that individuals lacking IgG2 should be regarded as agammaglobulinaemic and gammaglobulin replacement therapy initiated even if the total IgG is normal or even increased (Oxelius, 1979a). In addition to patients with selective IgG subclass deficiency, IgG2 subclass deficiency has also been described in increased frequency in a number of other immunodeficiency disorders such as IgA deficiency (Oxelius *et al.*, 1981), Ataxia-telangiectasia (Rivat *et al.*, 1969; Oxelius, 1979b; Rivat-Peran *et al.*, 1981) and cartilage hair syndrome (Oxelius, 1979b). In the latter diseases, the elevated risk for bacterial infections appear to be correlated to the lack of IgG2 rather than to the primary immunodeficiency. However, in this paper, a healthy blood donor (R.E.) lacking IgG2, IgA and IgE is described.

Relatively few studies have been made to date to quantitate the class and subclass distribution of antibodies to a specific antigen. The existing investigations have shown that antibodies to protein antigens are normally found in the IgG1 and IgG3 subclasses. These antigens include a variety of bacterial (Yount *et al.*, 1968; Carrel *et al.*, 1972; van der Giessen & Groenboer-Kempers, 1976), viral (Beck, 1981) and autologous (Natvig, Kunkel & Litwin, 1967; Tojo, Frion & Spiegelberg, 1970; Schur, Monroe & Rothfield, 1972; Karpatkin *et al.*, 1973; Perutz *et al.*, 1973; Lefvert & Bergström, 1977; Shakib & Stansworth, 1978) protein antigens. In contrast, anti-carbohydrate antibodies such as anti-dextran, anti-levan and anti-teichoic antibodies are restricted to the IgG2 subclass (Yount *et*

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0009-9104/83/0300-0600$02.00

al., 1968; Oxelius, 1974; Kraft *et al.*, 1982). Antibodies to a number of other bacterial polysaccharides also appear to be limited to the IgG2 subclass (Oxelius, 1974; Riesen, Skvaril & Braun, 1976; Siber *et al.*, 1980). Although isohaemagglutinins and heteroagglutinins have been suggested to be mainly of the IgM class, these antibodies also appear to be missing in IgG2 deficient patients (Oxelius, 1974), suggesting a functional linkage between anti-carbohydrate antibodies of the two different immunoglobulin classes.

Since individuals with IgG subclass deficiency offer a unique possibility to determine the subclass distribution of specific antibodies, the serum titres of the IgG2 deficient blood donor described in this paper were assayed for a variety of antigens.

CASE HISTORY

R.E. was born in 1944 after a normal pregnancy. In his childhood years he was vaccinated against Poliovirus, Diphteria, Tetanus toxoid and small pox. During these years he did not suffer from otitis, pneumonia or sinuitis, nor was he unduly susceptible to GI or viral infections. In his adult years, he had had infrequent viral upper respiratory tract infections. He has never received gammaglobulin injections or blood transfusions. In 1973, when routinely checked as a blood donor, no IgA could be found in his serum and he has since served regularly as a plasma donor.

MATERIALS AND METHODS

Cells. Lymphocytes were prepared from heparinized blood by floatation on Lymphoprep (Nyegaard A/S, Oslo, Norway) and cultivated and assayed for immunoglobulin secretion according to previously published methods (Hammarström *et al.*, 1979). Granulocytes were prepared by dextran sedimentation and then tested for functional capacity according to standard methods (Casciato *et al.*, 1975; Babior, 1978).

Immunoglobulin quantitations. Levels of IgM, IgG, IgA and IgD were measured in commercial immunodiffusion plates (Behringewerke, Marburg, West Germany). IgG subclasses were kindly measured by Dr Vivianne Oxelius (Department of Pediatrics, Lunds University Hospital, Lund, Sweden). IgE levels were kindly measured in a radioimmunoassay by Dr Ulla Persson (Department of Clinical Immunology, Karolinska Hospital, Stockholm, Sweden). Anti-blood group antigen antibodies and anti-sheep red cell antibodies were determined by passive haemagglutination. Anti-dextran antibodies were measured according to the method of Hedin & Richter (1982). Anti-viral and anti-bacterial titres, determined by complement fixation, haemagglutination inhibition assays or by newly developed ELISA techniques, were kindly measured by Professor Britta Wahren and Dr Marta Granström at the State Bacteriological Laboratory, Solna, Sweden according to previously published methods (Sundkvist & Wahren, 1981; Granström *et al.*, 1982).

Tissue typing was performed as described earlier in a microlymphocytotoxic method (Kissmeyer-Nielsen & Kjerbye, 1976; Bodmer, 1977).

RESULTS

Immunoglobulin quantitations

During routine screening of IgA deficient blood donors, a healthy male donor (R.E.) was found lacking IgA and IgE and with a low level of IgG (Table 1). Electrophoresis showed a typical pattern suggesting IgG2 deficiency (absence of anodal component of gammaglobulins) and when tested in electroimmunoassay, IgG2 was found to be virtually absent (0·03 g/l, normal adult levels 1·17–7·47 g/l). The κ/λ ratio was normal when tested in immunodiffusion. Upon repeated testing, no secretory IgA could be found in the saliva ($< 0{\cdot}0002$ g/l). Immunoglobulin levels in R.E.s two children and wife were normal although both children had inherited the putatively IgA deficiency predisposing HLA-A1, B8, DR3 haplotype from the father (Hammarström & Smith, 1982) (both children being

Table 1. Immunoglobulin quantitations

Donor	Age	Immunoglobulin levels IgM*	IgG	IgA	IgD	IgE†
R.E.	37	0·34	5·6	<0·02	<0·06	<0·125
K.E.	36	1·9	11·2	1·9	0·1	37·8
S.E.	12	1·4	12·4	1·3	<0·06	3·2
F.E.	7	0·6	8·9	0·7	<0·06	16·8
Normal range		0·3–2·5	7–16	0·5–3·3	≤0·1	1·6–122

* Given in g/l; †given in kU/l.

HLA-A1,3,B8,35,Cw4,DR1,3). Surface immunoglobulin bearing cells were present in normal proportions in all family members although few (<0·2%) surface IgA positive cells were found in R.E.

Specific antibody titres

Anti-viral titres such as morbilli (1:20), CMV (IgM < 1:50, IgG 1:450), influenza B (1:5), polio 1,2 (1:800, 1:200) and 3 (1:10) and rubeola (1:8) were considered normal as were anti-bacterial titres such as pertussis, AS (100 U/ml) and aDNAs (100 U/ml). However, titres against teichoic acid and alpha-toxin (1:700 and 1:90) (normal age matched (16–60 years) control titres ($n=50$) 1:4,086 (s.e. = 312, range = 1:920–9,000) and 1:898 (s.e. = 45, range = 1:250–1,720) respectively) (Granström *et al.*, 1982) as well as anti-pneumococcal saccharide 19 (data not shown) were extremely low. Anti-blood group antigen titres were slightly lower than average (anti-A 1:4, anti-B 1:16, R.E. is blood group 0), as were anti-sheep red cell antibody titres (1:2). No anti-dextran antibodies could be found in passive haemagglutination. Complement levels were all normal as were levels of orosomucoid, CRP and alpha$_1$-anti-chymotrypsin.

In vitro *cell function*

Granulocyte function *in vitro* (NBT, chemotaxis using zymosan activated serum as attractant and phagocytosis/killing of *Staphylococcus albus* and *B. fragilis*) was normal. *In vitro* activation of lymphocytes with different mitogens (pokeweed mitogen and Epstein-Barr virus) did not result in formation of IgA secreting cells and the number of IgG plaque forming cells was consistantly (three experiments) lower than expected (data not shown).

DISCUSSION

In this paper, a healthy adult blood donor who lacks IgG2 is described. In addition, no IgA or IgE could be detected. Isolated IgG2 deficiency (Oxelius, 1974, 1979b) or combined deficiences of IgG2 and IgA (Oxelius *et al.*, 1981; Rivat *et al.*, 1969; Rivat-Peran *et al.*, 1981) have been suggested to ultimately result in an increased susceptibility to bacterial infections and when deficient patients are encountered, gammaglobulin replacment therapy should be initiated (for review see Oxelius, 1979b). However, it seems clear from the data reported in this paper, that compensatory mechanisms exist and that IgG2 deficiency will not necessarily result in disease susceptibility. These data are also supported by the findings reported by van Loghem *et al.* (1980), where a healthy adult IgG2 deficient donor is briefly mentioned. Furthermore, although their health status is not explicitly stated, a few IgG2 deficient relatives of hypogammaglobulinaemic patients have been described (van der Giessen *et al.*, 1976). In addition, ataxia-telangiectasia patients, although uniformly IgG2 deficient (Oxelius *et al.*, 1981), are not always unduly susceptible to bacterial infections, thus arguing against an obligatory role for IgG2 in the immune defence system. In fact, all subclasses of

IgG may occasionally be dispensable it seems since we recently encountered a healthy IgG deficient donor (IgG < 0·007 g/l).

Cases of selective immunoglobulin deficiencies offer a possibility to determine the subclass distribution of specific antibodies. In man, investigations addressing this question are rare. It appears from the available data that anti-carbohydrate antibodies are preferentially of the IgG2 subclass (Yount *et al.*, 1968; Oxelius, 1974; Riesen *et al.*, 1976; Siber *et al.*, 1980; Kraft *et al.*, 1982). Protein antigens on the other hand, mainly give rise to IgG1 and IgG3 (Natvig *et al.*, 1967; Yount *et al.*, 1968; Tojo *et al.*, 1970; Carrel *et al.*, 1972; Schur *et al.*, 1972; Karpatkin *et al.*, 1973; Perutz *et al.*, 1973; van der Giessen & Groenboer-Kempers, 1976; Lefvert & Bergström, 1977; Shakib & Stansworth, 1978; Beck, 1981) and, in the case of factor VIII, IgG4 (Anderson & Terry, 1968; Robboy *et al.*, 1970). These previous findings are confirmed in this paper, since extremely low titres of anti-teichoic acid, anti-dextran and anti-pneumocaccal polysaccharide antibodies were found in the IgG2 deficient donor whereas anti-protein antibody titres tested were largely normal with the exception of alpha-toxin. Thus, it seems as if though the repertoire of V genes, as expressed in the antibody repertoire, is different in each subclass. In this context, it is interesting to note the suggested lack of iso- and heteroagglutinins (normally IgM antibodies) in IgG2 deficient patients (Oxelius, 1974), a finding that clearly suggests a maturational association between the anti-carbohydrate antibodies of the two classes. However, isohaemagglutinins were present, albeit in a slightly lower titre than normal (which may possibly simply be due to the relatively low IgM level found in the serum), in the IgG2 deficient donor described in this paper. In addition, in the serum of a IgG2 deficient patient (kindly provided by Dr Anders Freijd, Department of Oto-Rhino-Laryngeology, Huddinge University Hospital, Huddinge, Sweden), normal isoagglutinin titres were found (unpublished data), thereby strongly arguing against a functional linkage between IgM and IgG2 anti-carbohydrate antibodies. Furthermore, Yount *et al.* (1968) suggested that, although the antigen is of carbohydrate nature, isoagglutinins of the IgG class are in fact of the IgG1 subclass.

Thus, there appears not to be a strict IgG subclass dependency of anti-carbohydrate and anti-protein antibodies.

This work was supported by the Swedish Medical Research Council, the Petrus and Augusta Hedlunds Foundation, the Fredrik and Ingrid Thuring Foundation and the Walter, Ellen and Lennart Hesselman Foundation. The skilful technical assistance of Ms Silvia Nava, Ms Marie Schaffer and Ms Charlotte Åman is gratefully acknowledged. We are indebted to Dr Ulla Persson (Department of Clinical Immunology, Karolinska Hospital, Stockholm) for determination of IgE values and Professor Britta Wahren and Dr Marta Granström (State Bacteriological Laboratories, Solna) for measurements of anti-bacterial and anti-viral titres.

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