Release of Cytokines, Soluble Cytokine Receptors, and Interleukin-1 Receptor Antagonist After Intravenous Immunoglobulin Administration In Vivo

By Pål Aukrust, Stig S. Frøland, Nina-Beate Liabakk, Fredrik Müller, Ingvild Nordøy, Charlotte Haug, and Terje Espevik

We investigated the in vivo effects of one bolus injection (400 mg/kg) of intravenous immunoglobulin (IVIG) on a number of cytokines, soluble cytokine receptors, and interleukin-1 receptor antagonist (IL-1Ra) in plasma in 12 patients with primary hypogammaglobulinemia. A significant and rapid increase in plasma levels of IL-6, IL-8, and tumor necrosis factor α (TNFα) was seen within 1 hour after IVIG infusion. This increase was accompanied by a more prolonged elevation in levels of both types of soluble TNF receptors (sTNFRs), which remained elevated throughout the study period (44 hours) although they reached peak levels within 1 hour. After an initial rate in the ratio between TNFα and sTNFRs, this ratio decreased to values significantly lower than baseline values 20 and 44 hours postinfusion with ~600-fold molar excess of sTNFRs to TNFα (trimer). Although only a modest but statistically significant increase in plasma levels of IL-1α was seen, IVIG infusion was followed by a marked increase in plasma levels of IL-1α with 1,000-fold molar excess of IL-1Ra to IL-1α in some patients. The demonstrated effects of IVIG infusion on the cytokine network, particularly the induction of IL-1Ra and sTNFRs release, might be important for the therapeutic effects of IVIG in several immune-mediated disorders.

© 1994 by The American Society of Hematology.

TREATMENT WITH intravenous immunoglobulin (IVIG) is used not only for patients with primary or secondary hypogammaglobulinemia, but also increasingly in a number of noninfectious immune-mediated disorders (eg, autoimmune thrombocytopenic purpura, myasthenia gravis, Kawasaki disease, and systemic lupus erythematosus).1,2 Although data from several trials suggest its effectiveness in the modulation of abnormal cytokine production may contribute to the therapeutic effects of IVIG in these diseases,2,3 Several nonmutually exclusive mechanisms have been postulated, eg, blockade of Fc-receptor function of phagocytic cells, inhibition of production and/or neutralization of pathogenic autoantibodies possibly mediated by anti-idiotypic antibodies, changes in distribution and function of T-cell subsets, and antigen-specific immune suppression through antimicrobial or antiautoantibodies.1,2

Altered cytokine production may be involved in the pathogenesis of several autoimmune disorders,4 and therefore, modulation of abnormal cytokine production may contribute to the therapeutic effects of IVIG in these diseases.5,6 In vitro studies suggest that IVIG has direct effects on cytokine production in T-cells and monocyte/macrophages.3,4 However, in vitro experiments may not reflect the much more complicated situation in vivo, and very few studies have addressed the influence of IVIG on circulating cytokine levels.8 Furthermore, accumulating evidence indicates that biologic effects of a cytokine in vivo may be modulated not only by its soluble receptors and receptor antagonist, but also by other cytokines.9,10 Therefore, in the present study, we investigated the effects of IVIG on plasma concentrations of a number of cytokines, soluble cytokine receptors, and interleukin-1 receptor antagonist (IL-1Ra) in patients with primary hypogammaglobulinemia.

MATERIALS AND METHODS

Study population. Twelve patients (4 men and 8 women; median age, 39 years; range, 20 to 66 years) under treatment at the Section of Clinical Immunology and Infectious Diseases (Medical Department A, The National Hospital, Oslo, Norway) with the diagnosis of primary hypogammaglobulinemia based on established criteria12,13 were included in the study. Ten patients were classified as common variable immunodeficiency and two as congenital hypogammaglobulinemia as previously described.14,15 All patients had been treated with subcutaneous self-administered Ig for a minimum of 15 months and all had serum IgG levels greater than 5.0 g/L before the study. No patients had shown any signs of overt infection during the last 3 weeks before blood collection. None were taking antibiotics or immunosuppressive drugs except for one patient who received tetracyclines because of rosacea. At the time of the study, serum level of alamine aminotransferase was greater than 55 U/L and serum creatinine level less than 100 μmol/L in all patients.

Ig preparation. Octagam (Octapharma, Vienna, Austria) is a liquid virus-inactivated IVIG preparation (pH 4) produced from Norwegian fresh frozen plasma collected in Norwegian blood banks. The final product is dispensed in sterile water containing 10% maltose (final IgG concentration, 5 g/L; IgA and IgM, less than 0.1 g/L). Each portion has been tested and found negative for antibodies to human immunodeficiency virus and hepatitis B and C virus. The endotoxin level in the IVIG preparation was less than 10 pg/mL (limulus amebocyte lysate test).

Study design. The study was part of an Octagam tolerance and half-life study performed at Section of Clinical Immunology and Infectious Diseases. When entering the trial, the patients underwent a careful clinical, hematologic, and biochemical investigation. All patients received a single infusion of Octagam (0.4 g/kg) by slow intravenous infusion using an infusion set with filter (median total infusion time 3.2 hours; range, 2.7 to 4 hours). The patients were clinically followed during infusion and until 6 hours postinfusion.

© 1994 by The American Society of Hematology.

From the Section of Clinical Immunology and Infectious Diseases, Medical Department A, Kaptein W. Wilhelmsen og frues Institute of Bacteriology, University of Oslo, The National Hospital, Rikshospitalet, Oslo; and UNIGEN Center for Molecular Biology, Institute of Cancer Research, University of Trondheim, Trondheim, Norway.

Submitted March 9, 1994; accepted May 25, 1994.

Supported by Octapharma, Hurdal, Norway; Norwegian Research Council for Science and the Humanities: Anders Jahre’s Foundation; Medisnova Foundation; and Odd Kåre Rabben’s Memorial Fund for AIDS research.

Address reprint requests to Pål Aukrust, MD, Section of Clinical Immunology and Infectious Diseases, Medical Department A, Rikshospitalet, N-0027 Oslo, Norway.

The publication costs of this article were defrayed in part by page charge payment. This article must therefore be hereby marked "advertisement" in accordance with 18 U.S.C. section 1734 solely to indicate this fact.

© 1994 by The American Society of Hematology.

0006-4971/94/8407-0020$3.00/0

Blood, Vol 84, No 7 (October 1), 1994; pp 2136-2143

2136
Blood samples for plasma collection were taken preinfusion and postinfusion 1, 3, 20, and 44 hours. The study was approved by the Regional Ethical Committee and by the Norwegian Medicines Control Authority (Statens Leggemiddelkomite, SLK). Signed informed consent was obtained from each patient before entering the study.

**Blood sampling protocol.** Blood was drawn into sterile pyrogen-free vacuum blood collection tubes (Sarstedt, Numbrecht, Germany) using heparin (15 U/mL blood) as anticoagulant. Blood was immediately immersed in melting ice and the tubes were centrifuged within 20 minutes (400g and 4°C for 10 minutes). Plasma was then transferred in sterile Eppendorf tubes (Treff AG, Degersheim, Switzerland) and further centrifuged at 10,000g and 4°C for 5 minutes to obtain platelet-free plasma. Plasma was stored at -70°C in multiple aliquots until analysis (median storage time, 10 weeks; range, 1 to 22 weeks). Samples were frozen and thawed only once. To minimize the variability, all samples from a given patient were run at the same time (on the same microtiter enzyme immunoassay [EIA] plate).

**Immunoenzymometric assay for detection of soluble tumor necrosis factor receptors (TNFRs) in plasma.** The soluble TNFRs (sTNFRs), p55 and p75, were analyzed by immunoenzymometric assays described by Liabakk et al.18 Briefly, immunoplates were coated with the monoclonal antibodies (MoAbs) IV4E and 3H15, recognizing non-TNF-binding sites of p55- and p75 TNFR, respectively. Recombinant human (ru) p55- and p75 TNFR (provided by Dr H. Loetscher, F. Hoffmann-La Roche, Basel, Switzerland) served as standards. Bound sTNFRs were detected with digoxigenin (DIG)-labeled TNF, anti-DIG Fab fragments and o-phenylenediamine as substrate. The detection limit was 300 pg/mL for both receptor assays. The intraassay and interassay coefficients of variation were less than 10% for both assays. The specificity of the assays was controlled by inhibition with unlabeled ruTNFα together with DIG-labeled TNF. The combination of antibodies recognizing non–TNF-binding sites of sTNFRs with DIG-labeled TNF makes detection of free as well as reversibly occupied sTNFRs possible, because addition of excess DIG-labeled TNF displaces TNF bound in complex to sTNFRs. The presence of 10 ng/mL of TNFa in a sample did not influence the results, suggesting that the assay is not affected by the presence of TNFa in plasma samples even in pathologic conditions known to be associated with very high circulating TNFa levels (eg, severe meningococcemia).

**Other immunoenzymometric assays.** Plasma concentrations of cytokines, soluble IL-6Rs (sIL-6Rs), and IL-1Ra was quantified by EIAs provided from Medgenix (Fleurus, Belgium) (IL-1B, IL-6, TNFa, and interferon γ [IFNy]) and Quantikine, R & D (Minneapolis, MN) (IL-4, IL-8, IL-1α, sIL-6R) according to the manufacturer’s guidelines, using microtiter wells coated with one (Quantikine) or several (Medgenix) MoAbs against distinct epitopes of the actual cytokine. The recombinant IL-1Ra standard was provided from Synergen Inc (Boulder, CO). Samples with high levels of TNFa or IL-1Ra were diluted and reassayed, and in the sIL-6R EIA, all samples were run in two dilutions (1/50 and 1/100). The IL-6 and TNFa EIAs detect free IL-6 and TNFa as well as these cytokines bound to their soluble receptors17 (manufacturer’s information). There was no cross-reactivity in the EIAs between the measured parameters or with other known cytokines or soluble cytokine receptors (manufacturer’s information). In all EIAs, samples and recombinant standards (n = 7) were run in duplicate, and the optical density (OD) measured in a Multiskan Multisoft photometer (Labsystems, Helsinki, Finland). The standard curve for each plate was obtained by fitting the average OD readings for the standards to a third order polynomial, and the concentrations of the samples were determined from the obtained standard curve (ELISA+ software, Meddata Inc, New York, NY). According to the manufacturer’s instructions, the lower detection limits of the EIA kits were 2 pg/mL (IL-1β), 4.1 pg/mL (IL-4), 3 pg/mL (IL-6), 18.1 pg/mL (IL-8), 0.03 IU/mL (IFNy), 3 pg/mL (TNFa), 22.0 pg/mL (IL-1Ra), and 350 pg/mL (sIL-6R, when assayed in a 100-fold dilution), and at our laboratory, the lowest standard was used to equal the detection limit. The intraassay and interassay coefficients of variation for all assays were less than 10%.

**Statistical analysis.** For comparison of plasma concentrations after IVIG infusion with baseline levels the nonparametric Wilcoxon Matched Pairs Test was used. Coefficients of correlation (r) were calculated by the Spearman rank test. The calculations were performed using the Statistical Analysis System (SAS Institute Inc, Cary, NC) and STATISTICA/Mac (StatSoft, Tulsa, OK) software packages. In the text, peak level is defined as the highest measured absolute concentration, and maximum change is defined as the measured concentration deviating maximally from baseline level – baseline concentration. Data are given as medians and 25th to 75th percentiles if not otherwise stated. P values are two sided and considered significant when P < .05.

**RESULTS**

**Cytokine levels in IVIG preparation.** Using the EIAs described above, we could not detect any amount of IL-1β, IL-4, IL-6, IL-8, TNFa, IFNy, IL-1Ra, or sTNFRs in the IVIG product used during the study. Soluble IL-6R could be detected in very low concentrations (510 pg/mL), well below both the lowest baseline value in the patients (25.5 ng/mL) and the lowest detectable level in 40 healthy blood donors (13.5 ng/mL, manufacturer’s information).

**Cytokine responses after IVIG infusion.** As shown in Fig 1, there was a significant increase in plasma levels of IL-6, IL-8, and TNFa after IVIG administration. All patients had an increase in circulating IL-8 level and all but one, who had a markedly elevated baseline level of TNFa, had an increase in circulating IL-6 and TNFa levels. However, the kinetics of these cytokine changes were somewhat different (Fig 1). Thus, 7 of 12 patients reached peak level for IL-6 after 1 hour, whereas IL-8 peaked in nine patients 3 hours postinfusion. Although the highest median TNFa level was reached 3 hours postinfusion, seven patients, including those two with the most pronounced increase in plasma level, reached peak level after 1 hour. After 3 hours, there was a decline in circulating levels of these cytokines, but plasma levels of IL-6 and IL-8 in contrast to TNFa concentrations were still significantly increased compared with baseline values after 20 hours (Fig 1).

There was a slight, but significant increase in circulating IL-1β level (0 pg/mL [0 to 0 pg/mL] versus 4 pg/mL [0 to 7 pg/mL], P < .05, baseline and 3 hours postinfusion, respectively). However, five patients had undetectable plasma levels throughout the study.

Although there was a slight increase in the median IFNy level after infusion, the differences from baseline values were not statistically significant (Fig 1). IVIG infusion did not induce any significant changes in plasma level of IL-4, and most patients had undetectable plasma levels throughout the study (data not shown).

**IL-1Ra response after IVIG infusion.** As illustrated in Fig 1, there was a marked increase in circulating IL-1Ra concentration after IVIG infusion. All patients had an increase in plasma levels with peak level at 1 hour postinfusion (11 of 12 patients). IL-1Ra levels returned rapidly to baseline values (Fig 1).

**Plasma levels of soluble cytokine receptors after IVIG**
infusion. In contrast with circulating IL-1Ra, plasma levels of sTNFRs and sIL-6R showed a more prolonged elevation after IVIG infusion (Fig 1). Soluble TNFRs showed a significant and rapid increase in circulating levels with peak values at 1 hour postinfusion for 7 of 12 and 6 of 12 patients, p55 and p75 receptors, respectively, and plasma concentrations remained elevated throughout the observation period (44 hours) (Fig 1). Only 2 patients did not show an increase in plasma levels for both types of sTNFRs (1 with markedly increased baseline levels for both sTNFRs, and 1 with an increase only in p75 level after IVIG infusion).

There was a persistent, but not significant increase in plasma levels of sIL-6R after IVIG infusion (Fig 1) and except for the two patients with the highest baseline concentrations, an increase was observed in all patients.

**Balance between TNFα and sTNFRs after IVIG infusion.** The ratio between TNFα and the sTNFRs may provide an estimate of the balance in plasma between TNF molecules and sTNFRs. In molecular terms, this ratio was defined as immunoreactive TNFα (pmol/L)/(p55 + p75)(pmol/L) × 100, assuming a molecular mass of 17 × 3 kD and 30 kD for TNFα (trimer) and both types of sTNFRs, respectively. Thus, a ratio less than 0.2 reflects a more than 500-fold molar excess of sTNFRs to TNFα (trimer). When analyzing patients with a detectable TNFα level at baseline (n = 7), there was a significant increase in this defined ratio at 1 hour postinfusion (Fig 2). However, at 20 and 44 hours postinfusion, a significant decrease was observed compared with baseline values, with ~600-fold molar excess (median) of sTNFRs at 44 hours postinfusion (Fig 2).

**Correlations between plasma levels of cytokines and their soluble cytokine receptors after IVIG infusion.** As shown in Table 1, the maximum changes in plasma level of TNFα were strongly positively correlated with the maximum changes in plasma levels of both types of sTNFRs. Moreover, the increase in p75 and p55 concentrations was significantly positively intercorrelated (Table 1). However, when analyzing the correlations between TNFα concentration and sTNFRs levels at various time points, only p75 levels were significantly correlated with the TNFα values (Table 2).

In contrast with the strong positive correlation between maximum changes in TNFα and sTNFRs levels, no significant correlation was found between IL-6 and sIL-6R (Table 1).

**Correlations between changes in plasma levels of IL-1Ra and other parameters.** The maximum change in IL-1Ra level was positively correlated with the maximum changes in all measured TNF parameters (Table 1). No significant correlation was found between maximum changes in IL-1Ra and IL-1β concentrations (data not shown).

**Correlations between plasma levels of different cytokines after IVIG infusion.** The maximum changes in TNFα plasma level tended to be positively correlated with the maximum changes in IL-6 and IL-8 concentrations (Table 1). In contrast to TNFα, increase in IFNγ plasma level seems to be negatively correlated with increases in the other measured parameters, and this negative correlation was statistically significant for IL-8 and sIL-6R (Table 1).

**DISCUSSION**

The present study clearly shows that IVIG administration in vivo may have a number of effects on the cytokine network as reflected in plasma concentrations of cytokines and soluble cytokine receptors. Thus, IVIG infusion was followed by a significant increase in proinflammatory cytokines (eg, IL-8 and TNFα) as well as cytokine mediators with
anti-inflammatory properties (eg, IL-1Ra, sTNFRs, and IL-6).

Several effects of IVIG administration were seen on the various components of the TNFα system. Soluble TNFRs are released from different cell types after proteolytic cleavage of the membrane TNFRs. The mechanisms controlling TNFRs shedding have not been fully elucidated. However, it has been suggested that both TNFα and stimuli causing TNFα release also induce shedding of sTNFRs. Thus, sTNFRs in plasma/serum are thought to indicate activation of the TNFα system with elevated concentration persisting for a longer time than TNFα after stimulation. In the present study there was a significant positive correlation between the maximum change in TNFα and the levels of both types of sTNFRs after IVIG infusion. Others have previously shown a positive correlation between peak levels of these parameters in patients with sepsis and in patients with severe malaria. Furthermore, TNFα stimulation in vitro as well as in vivo has been shown to induce sTNFRs release. However, we observed that whereas plasma levels of soluble p75 receptor were significantly correlated to TNFα levels at baseline as well as at other time points, no significant correlation was shown between concentrations of soluble p55 receptor and TNFα at any time points, possibly reflecting differences in kinetics and regulation of these two types of sTNFRs, as has recently been suggested by others.

Although there was a statistically significant increase in plasma levels of IL-1β after IVIG infusion in the whole group, this increase was very modest. In contrast, IVIG infusion was followed by a marked increase in plasma levels of IL-1Ra, the structurally related, naturally occurring antagonist of IL-1. To our knowledge, the present report is the first published demonstration of induction of increased circulating IL-1Ra levels by IVIG infusion.

Although the correlation between the increase in IL-1Ra and soluble p75 receptor did not reach statistical significance, we found a positive correlation between the maximum change in IL-1Ra concentration and the maximum changes in TNFα and sTNFRs levels. Our findings may merely reflect correlation between different parameters of IVIG-induced monocyte hyperactivity because monocytes are a major source for both IL-1Ra and TNFα production. On the other hand, these correlations may reflect common inducing mechanisms for the release of these mediators after IVIG infusion. Interestingly, Mazzei et al have suggested that TNFα may be implicated in the IL-1Ra production in a promyelocytic cell line, possibly via induction of GM-CSF, and a very recent study has shown that TNFα greatly augmented IL-1Ra transcripts in neutrophils that may contribute to the circulating IL-1Ra levels.

We did not investigate the mechanisms of the cytokine modulating effects of IVIG administration in vivo. However, it is conceivable that some of the effects are mediated by interaction of immune complexes or IgG with Fc-receptors on monocytes, as well as other cell types, e.g., neutrophils and NK-cells. IgG prepared from pooled plasma may spontaneously dimerize by anti-idiotypic interactions and antigens present in the body may form immune complexes with antibodies in IVIG. Previous in vitro studies have shown that both immune complexes and IgG are potent stimulants of IL-1Ra production in monocytes without affecting IL-1β release. Immune complexes have also been shown to induce production of TNFα and IL-6 in monocytes and IFNγ in NK-cells via Fc-receptor interaction. Other mechanisms may involve cytokine antibodies and other immunomodulating factors (eg, sCD4, sCD8, sHLA antigens) in IVIG preparations. However, in the IVIG preparation used in the present study, we could neither detect sCD8 (unpublished

| Table 1. Correlations (r) Between Maximum Changes in Plasma Levels of Different Cytokines and Soluble Cytokine Receptors After IVIG Infusion |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| TNFα            | IL-6            | IL-8            | IFNγ            | p55 Receptor    | p75 Receptor    | sIL-6R          | IL-1Ra          |
| .47             | .47             | -.19            | .65*            | .73*            | -.03            | .63*            |
| -.11            | .19             | .68*            | .46             | .03             | .16             |
| -.88*           | .01             | .26             | .32             | .55             |
| .03             | -.16            | -.61*           | -.38            |
| Soluble p55 receptor | .75*          | -.44            | .63*            |
| Soluble p75 receptor | -.24          | .52             | -.08            |
| sIL-6R          |                 |                 |                 |                 |

* P < .05.
† P < .01.
‡ P < .002.

| Table 2. Correlations (r) Between Plasma Levels of TNFα and sTNFRs (p55 and p75 receptor) at Baseline and at Various Time Points After IVIG Infusion |
|-----------------|-----------------|-----------------|-----------------|-----------------|
| Correlation Between | Baseline | 1 h Postinfusion | 3 h Postinfusion | 20 h Postinfusion | 44 h Postinfusion |
| Soluble p55 receptor | .44            | .38             | .55             | .36             | .26             |
| Soluble p75 receptor | .85*           | .781            | .83*            | .741            | .631            |

* P < .002.
† P < .01.
‡ P < .05.
The demonstrated effects of IVIG on the circulating cytokine network may be relevant for the clinically significant immunomodulating effects of IVIG therapy seen in many disorders. The proinflammatory cytokines IL-1 and TNFα play an important role in many autoimmune diseases, and therefore, suppression of the activity of these cytokines may be an important therapeutic goal in such disorders. Although there was an initial increase in the plasma levels of TNFα and IL-1β, this increase was accompanied by an increase in levels of both types of sTNFRs as well as IL-1Ra. Furthermore, plasma levels of sTNFRs remained persistently elevated during the whole study period of 44 hours in contrast with TNFα levels. Soluble TNFRs may neutralize TNFα in vitro and sTNFRs have recently been shown to protect mice from lethal effects of endotoxic shock. Based on such observations, it has been suggested that the degree of biologic TNFα activity may depend on the balance between TNFα and sTNFRs. In the present study, we observed that after an initial increase in the ratio between TNFα and sTNFRs, this ratio 20 and 44 hours postinfusion decreased to values significantly lower than baseline values with ~600-fold molar excess of sTNFRs to TNFα (trimer). It is conceivable that such a degree of sTNFRs excess may mediate an inhibition of TNFα-mediated cytokotoxicity.

Also, the increased levels of IL-1Ra found after IVIG infusion may be of clinical relevance. When comparing peak levels, a 1000-fold molar excess of IL-1Ra to IL-1β was found in some patients. A 10-fold molar excess of IL-1Ra is required for inhibition of IL-1-induced responses in monocytes in vitro, whereas IL-1Ra to IL-1β ratios greater than 10³ are needed to block IL-1 effects in vivo. Thus, the in vivo effects of the increased IL-1Ra levels after IVIG infusion are uncertain.

Increased levels of IL-6, IFNγ, and sIL-6R were observed after IVIG infusion, even if only the increase in IL-6 levels reached statistical significance. These changes may also be of some interest with reference to immunomodulating effects of IVIG therapy. Although IL-6 shares several biologic features with TNFα and IL-1, this cytokine also has anti-inflammatory properties. Thus, IL-6 has been shown to suppress lipopolysaccharide (LPS)-induced TNF and IL-1 production. Increased levels of sIL-6R might contribute to such anti-inflammatory effects because sIL-6R, in contrast to other soluble cytokine receptors, has agonistic properties together with its ligand. Furthermore, IFNγ may also exhibit anti-inflammatory properties and has been found to inhibit both IL-1 and IL-8 production in human monocytes and neutrophils, respectively. In the present study, the maximum changes in plasma levels of IFNγ and IL-8 were inversely correlated.

The results from the present study may seem at variance with some recent reports showing suppressive effects of IVIG on IL-6 and TNFα production in vitro. These discrepancies may have several explanations. First, Andersson et al and Shimosato et al investigated the suppressive effects of IVIG on IL-6 and TNFα production in peripheral blood mononuclear cells (PBMCs) by adding IVIG to cell cultures less than 1 hour before LPS stimulation, which might have made the cells refractory to further stimulation. Indeed, in another in vitro study, IVIG showed enhancing effects on TNFα production from PBMC when given alone and 24 hours before stimulation. Second, and even more important, cytokine production in vitro and in vivo differs considerably. In vivo, the measured circulating cytokine level is the outcome of the interplay between several cytokines, other circulating factors known to influence cytokine production (eg, hormones, neuropeptides), and a large numbers of cells (eg, PBMC, neutrophils, endothelial cells) not present in an in vitro system, making it very difficult to extrapolate results from simplified in vitro models to the much more complex in vivo situation.

The present study of the in vivo effects of IVIG has been performed in patients with primary hypogammaglobulinemia, whereas most studies examining the in vitro effects of IVIG have been performed on samples from healthy blood donors. Although the main clinical feature in these patients is recurrent bacterial infections, particularly in the respiratory tract, autoimmune disorders, noncaseating granulomata, lymphoid and gastrointestinal malignancies, and nonmalignant lymphoid hyperplasia are also common. We and others have previously shown that, in addition to the B-cell defect, many of these patients are characterized by a state of chronic immune activation involving both T-cells and monocyte/macrophages, which is reflected in raised serum levels of cytokines and other markers of immune activation. Therefore, it is conceivable that findings in these patients may have relevance to the clinical effects of IVIG in patients with autoimmune and other inflammatory disorders known to benefit from IVIG therapy. However, further studies in other patient populations will be needed to confirm and extend our findings.

It is probable that a number of different mechanisms are responsible for the clinical effects of IVIG therapy seen in many diseases with immune pathogenesis. We believe that the demonstrated effects of IVIG infusion on the cytokine network, particularly the induction of cytokines, soluble cytokine receptors, and cytokine receptor antagonist with anti-inflammatory properties, might be important for the therapeutic effects of IVIG in several immune-mediated disorders, at least for rapidly occurring effects observed within days after start of therapy.

ACKNOWLEDGMENT

We thank Lisbeth Wikeby and Bodil Lunden for excellent technical assistance and Tone Grande (Mericon, Skien, Norway) for helpful statistical consultation.

REFERENCES


10. Fernandez-Botran R: Soluble cytokine receptors: Their role in immunoregulation. FASEB J 5:2567, 1991


46. Schindler R, Mancilla J, Endres S, Ghorbani R, Clark SC, Dinarello CA: Correlations and interactions in the production of interleukin-6 (IL-6), IL-1, and tumor necrosis factor (TNF) in human blood mononuclear cells: IL-6 suppresses IL-1 and TNF. Blood 75:40, 1990


Release of cytokines, soluble cytokine receptors, and interleukin-1 receptor antagonist after intravenous immunoglobulin administration in vivo

P Aukrust, SS Froland, NB Liabakk, F Muller, I Nordoy, C Haug and T Espevik