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Monitoring of two intravenous immunoglobulin preparations for immunoglobulin G subclasses and specific antibodies to bacterial surface antigens and relation with their levels in treated immunodeficient patients

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Abstract

Patients with antibody deficiency disorders are highly susceptible to bacterial infections. Replacement therapy with intravenous immunoglobulin preparations (IVIG) has been established in such patients for two decades. The efficacy of IVIG treatment depends on the amount of functional pathogen-specific antibodies provided. The present study was undertaken to determine the levels of immunoglobulin classes, IgG subclasses, and specific antibodies to bacterial surface antigens in two different IVIG preparations (Sandoglobulin® and Gamimmune®) and blood sera of IVIG-treated immunodeficient patients. The levels of IgG, IgA, IgM and IgG subclasses were determined in both IVIG preparations and in patients' sera and were compared with those of healthy individuals. Sandoglobulin® contained significantly higher concentrations of IgA, IgG_2 and IgG_4 than Gamimmune®. The latter contained higher concentrations of IgG_1. Patients treated with Gamimmune® had significantly lower concentration of IgG_4 as compared with healthy individuals and Sandoglobulin®-treated patients. This finding was related to the preparation's composition. Screening of 20 lots from each preparation for antibodies to frequent clinically isolated strains of *Escherichia coli*, *Staphylococcus aureus*, *S. epidermidis*, *Klebsiella pneumoniae* and *Enterococci* spp. showed a high lot-to-lot variability. In order to overcome the lot-to-lot variability and correlate the observed effects with each IVIG preparation, the administered IVIG lots were selected so that their titers were in the interval of mean value $\pm$ S.D. for each pathogen. The two tested preparations showed significant differences in their content of specific antibodies that ultimately affected the levels of these antibodies in treated patients. More specifically, Sandoglobulin® contained higher levels of antibodies to *E. coli* and *S. epidermidis* strains. Infusion of this preparation maintained the respective antibodies in the recipients significantly higher than those of healthy individuals. Gamimmune® infusion led to similar

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and comparable levels. Both IVIG preparations had comparable antibody titers towards *K. pneumoniae*, provided high amounts of antibodies, and kept recipients' specific IgG at levels significantly higher than those of the healthy individuals. *Enterococci* spp. specific antibodies were significantly higher in Gamimmune®, whereas titers of antibodies towards *S. aureus* were comparable. Levels of antibodies against both *Enterococci* spp. and *S. epidermidis* after administration of both preparations were close to those in healthy individuals. None of the patients developed infection during the time of the study. In conclusion, most of the lots of the two IVIG preparations studied, despite some quantitative differences, provide patients with sufficient amounts of antibodies to bacterial surface antigens that protect them against infections.

Keywords: Enzyme immunoassay; Intravenous immunoglobulin; Pathogenic bacteria; Primary immunodeficiency

1. Introduction

Intravenous human polyvalent immunoglobulin (IVIG) has successfully replaced intramuscular preparations used since 1952 in the treatment of primary and acquired immune deficiencies [1–4]. Patients with primary antibody deficiency disorders are highly susceptible to frequent and severe infections caused by common bacterial pathogens. Administration of IVIG may reduce infection rates when the preparation maintains serum IgG at high levels providing adequate quantities of antibodies specific to infectious organisms.

It has been demonstrated that IVIG contains significant titers of antibodies to various pathogenic bacteria [5]. We have recently developed an enzyme-linked immunosorbent assay (ELISA) to determine antibodies specific to bacterial surface antigens in IVIG preparations [6]. Titers of antibodies toward clinically isolated Gram negative strains significantly differed not only between different IVIG preparations but also among lots from the same preparation [6]. Other studies have clearly shown that there is good correlation of antibody titers and opsonic activity [7,8], as well as their protection capacity in animal models [9]. The aim of this study was: (1) To investigate the composition of two commercially available IVIG preparations in terms of immunoglobulin classes, IgG subclasses, and antibodies specific to clinically isolated bacterial pathogens. (2) To determine the same parameters in patients with primary antibody deficiency states after infusion of each IVIG preparation in order to study the relationship between the IVIG composition and the efficacy of IVIG treatment.

2. Experimental

2.1. Patients and clinical protocol

Patients with primary immunodeficiencies ($n = 10$, males, 5–21 years old) received Sandoglobulin® (7S intact IgG isolated by precipitation at pH 4 and pepsin treatment, Novartis, Switzerland) and Gamimmune® (7S intact IgG isolated by diafiltration at pH 4.25, Bayer, US) as replacement therapy in alternate months. Particularly, Sandoglobulin® (6% w/v, pH 6.9) and Gamimmune® (5% v/v, pH 4.25) were infused (0.5–1 g kg^{-1} body wt. per 30 days) to all patients at the same rate (2.5 ml min^{-1}). Five of the patients suffered from X-linked agammaglobulinaemia and five from common variable immunodeficiency. All patients included in this study had been continuously treated with Sandoglobulin® once every 30 days before the initiation of the present study for a period varying from 2 to 20 years (long-period treated group). Sera were collected before IVIG administration, 30 days after Sandoglobulin® infusion and 30 days after Gamimmune® administration. Healthy individuals ($n = 10$, males, 5–22 years old) comprised the control group. The levels of IgG, IgA and IgM as well as the subclasses of IgG were determined in all IVIG lots used for the infusions and in serum samples using rate nephelometry (Behring Nephelometer Analyser).

2.2. Bacterial isolates and materials

Gram positive and negative bacteria (*Klebsiella pneumoniae*, *Enterococci* spp., *Escherichia coli*, *Staphylococcus aureus* and *S. epidermidis*) were

isolated from patients hospitalized in the Intensive Care Unit at the University Hospital of Patras. Stock cultures of all strains were maintained in Tryptic Soy Broth (BBL Microbiology, Cockeysville, MD) supplemented with 20% glycerol at −70° without serial passage. Peroxidase-H conjugated rabbit anti-human IgG and *ortho*-phenylenediamine tablets of 1 mg were obtained from Sigma (St Louis, MO). All other chemicals used were of analytical grade.

2.3. Determination of specific antibodies toward bacterial surface antigens

Enzyme-linked immunosorbent assay (ELISA) was performed on sterile 96-well round-bottomed microplates as previously described [6]. Briefly, suspensions of various bacterial species with an original optical density 1.0 at 600 nm were diluted 1:30 with phosphate-buffered saline (PBS). The plates were coated with the suspensions (100 μl $well^{-1}$) at 4°C for 16 h and were washed with PBS containing 0.1% Tween 20, 0.4 N NaCl and 0.5% (w/v) bovine serum albumin (BSA) (PBS–Tween) (150 μl $well^{-1}$). Incubation with a 3% (w/v) solution of BSA in PBS (200 μl $well^{-1}$) at 37°C for 1 h served to prevent non-specific binding. Microplates were washed three times with PBS–Tween, and incubated with various dilutions of IVIG or blood sera at 37°C for 1 h. Following three washings, peroxidase H-conjugated rabbit anti-human IgG, diluted 1:4000 with PBS, was used as detection antibody and was incubated at 37°C for 1 h. Color developed by adding *ortho*-phenylenediamine at 0.067% (w/v) in 0.1 M sodium citrate acid in the presence of 0.03%(v/v) H_2O_2 as enzyme substrate. The mixture was incubated for 15 min at room temperature in the absence of light. The reaction was terminated with 1 M H_2SO_4 and the optical density was measured at 490 nm in a Molecular Devices E-max photometer. As a control of non-specific binding of the preparations, microplates were coated only with BSA. Obtained blank values were always automatically subtracted from those obtained in the samples.

Absorbance at 490 nm (A_{490}) was plotted against the sample dilution in a log–log diagram. The dilution of IVIG or sera giving an A_{490} of 0.2 above the background was multiplied by the IgG concentration in g ml^{-1} and the resulting value in U ml^{-1} was the titer of pathogen-specific antibodies [6]. Calibration and validation of results were performed using the SOFT max PRO software (version 1.2.0).

3. Results

3.1. Variations of pH and immunoglobulin levels in IVIG preparations and patients' sera

During the study period, none of the patients showed clinical signs of infection. Both IVIG preparations were well tolerated by patients and adverse-reactions were not recorded either at the time of infusion or later. The pH values of the preparations (pH 6.38 ± 0.04 for Sandoglobulin® and pH 3.9 ± 0.069 for Gamimmune®) were significantly different ($P \leq 0.001$). However, screening of pH values in healthy individuals and patients' blood sera before, during, and after infusion of the two IVIG preparations revealed no significant differences.

The levels of immunoglobulin classes (IgG, IgA and IgM) and IgG subclasses (IgG_1, IgG_2, IgG_3 and IgG_4) in the two IVIG preparations and immunodeficient patients before and after IVIG administration are presented in Tables 1 and 2, respectively. As indicated in Table 1, the two IVIG preparations contained IgG as the major immunoglobulin class in comparable levels. Although low levels of IgA were identified, IgA content in Sandoglobulin® was significantly higher ($P \leq 0.0001$) than that in Gamimmune®. Three out of four IgG subclasses were found to differ between the two IVIG preparations. Thus, Sandoglobulin® contained significantly ($P \leq 0.0001$) higher levels of IgG_2 and IgG_4, whereas Gamimmune® had higher levels of IgG_1 ($P \leq 0.01$).

As shown in Table 2, total serum IgG concentration in all IVIG treated patients was slightly lower than that of healthy individuals but no statistical difference was recorded. Intravenous immunoglobulin substitution therapy, therefore,

Table 1
Content of immunoglobulin classes and IgG subclasses in IVIG preparations[a]

	Sandoglobulin®	Gamimmune®
Immunoglobulin classes		
IgG	53.53 ± 5.49	57.48 ± 3.19
IgA	0.99 ± 0.12*	0.09 ± 0.02
IgM	<0.04	<0.04
IgG subclasses		
IgG_1	27.43 ± 3.17	31.60 ± 0.86**
IgG_2	14.70 ± 1.20*	10.61 ± 0.45
IgG_3	2.04 ± 0.16	2.22 ± 0.16
IgG_4	1.99 ± 0.30*	0.34 ± 0.05

[a] Results are expressed as g l^{-1}. Values are mean ± S.D.
* Significantly higher values ($P \leq 0.001$) between the two IVIG preparations.
** Significantly higher values ($P \leq 0.01$) between the two IVIG preparations.

was considered successful since it maintained average serum IgG at preinfusion levels well above the critical value of 7.0 g l^{-1}.

IgA levels in all IVIG treated patients were significantly ($P \leq 0.001$) lower than those in healthy individuals. Although the levels of IgA in Sandoglobulin® (0.99 ± 0.12 g l^{-1}) were significantly higher than those in Gamimmune® (0.09 ± 0.02 g l^{-1}), this difference did not affect IgA levels in blood sera of IVIG-treated patients since no significant differences were observed following either Sandoglobulin® or Gamimmune® infusion. IgM could not be detected in patients' blood sera by the analytical methods used, since their levels were lower than the detection limit (0.04 g l^{-1}).

IgG_1 levels in all IVIG-treated patients were significantly ($P \leq 0.05$) lower than those in healthy individuals (Table 2). Although IgG_1 concentration of Gamimmune® was higher ($P \leq 0.01$) than that of Sandoglobulin®, no significant differences were found following administration of the two IVIG preparations. IgG_2 levels in immunodeficient patients were comparable with those in healthy ones. Although the two preparations differed significantly ($P \leq 0.0001$) in IgG_2 content, their administration resulted in similar IgG_2 levels in patients' sera. IgG_3 levels in long-period treated patients were slightly lower than those in the healthy group, but no statistically significant differences between them were recorded. Administration of both IVIGs resulted in comparable serum IgG_3 levels. Obtained levels were also slightly lower as compared with those of healthy individuals. IgG_4 levels in long-period

Table 2
Content of immunoglobulin classes and IgG subclasses in healthy individuals and immunodeficient patients before and after IVIG administration[a]

	Healthy individuals	Long-period treated patients[b]	After Sandoglobulin®[c]	After Gamimmune®[d]
Immunoglobulin classes				
IgG	10.27 ± 1.86	7.85 ± 1.60	8.64 ± 2.08	7.91 ± 1.43
IgA	1.25 ± 0.30	0.37 ± 0.23*	0.38 ± 0.29*	0.38 ± 0.25*
IgM	1.16 ± 0.27	ND[e]	ND	ND
IgG subclasses				
IgG_1	6.01 ± 0.92	4.37 ± 1.25*	4.59 ± 1.34*	4.19 ± 0.96*
IgG_2	2.35 ± 0.84	2.47 ± 0.65	2.64 ± 0.61	2.32 ± 0.48
IgG_3	0.32 ± 0.08	0.26 ± 0.14	0.28 ± 0.17	0.26 ± 0.19
IgG_4	0.41 ± 0.18	0.34 ± 0.14	0.32 ± 0.12	0.16 ± 0.13*,**

[a] Results are expressed as g l^{-1}. Values are mean ± S.D.
[b] Levels in patients with primary immunodeficiencies treated with Sandoglobulin® for a long period (2–20 years).
[c] Levels in patients 30 days after Sandoglobulin® infusion.
[d] Levels in patients 30 days after Gamimmune® infusion.
[e] ND, not detected.
* Significant differences ($P \leq 0.05$) between healthy individuals and patients.
** Significant differences ($P \leq 0.05$) between groups of treated patients following IVIG administration for a 30-day period.

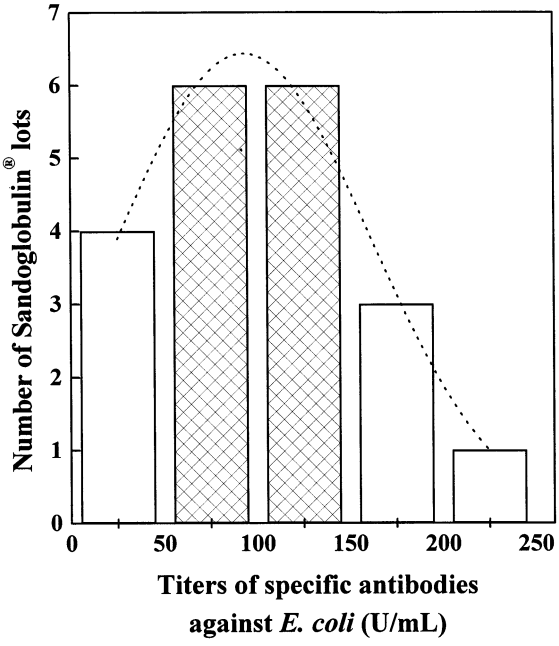

Fig. 1. Histogram depicting the frequency of Sandoglobulin® lots with titers of *E. coli*-specific antibodies within an interval of 50 U ml^{-1}. Marked bars represent the lots which are within the interval mean ± S.D. and were finally selected for administration.

Table 3
Titers of specific antibodies toward bacterial antigens in selected lots of the two IVIG preparations[a]

Bacteria	Sandoglobulin®	Gamimmune®
E. coli	104.27 ± 15.937*	ND[b]
S. aureus	150 508 ± 69 050	101 880 ± 52 810
K. pneumoniae	18.675 ± 2.97	20.64 ± 3.31
S. epidermidis	191.97 ± 28.41*	134.89 ± 20.25
Enterococci spp.	5.55 ± 0.83	12.43 ± 1.89*

[a] Results are expressed in U ml^{-1} estimated as previously described [13]. Values are mean ± S.D. of nine lots in triplicate.

[b] ND, not detected.

* Statistically ($P < 0.001$) higher titers between the two IVIG preparations.

treated patients were comparable with those of healthy individuals. Gamimmune®, which contained significantly lower ($P < 0.0001$) levels of IgG_4, was not capable of maintaining the levels as high as those obtained by Sandoglobulin®. Particularly, IgG_4 levels following Gamimmune® infusion were found to be significantly ($P \leq 0.05$) lower by 27% as compared with those of Sandoglobulin®-treated patients and healthy individuals.

3.2. Titers of antibodies toward bacterial surface antigens in IVIG and blood sera

The content of antibodies against certain bacterial isolates was determined in both IVIG preparations, in healthy individuals, and in patients' sera before and after IVIG administration using an ELISA method [6]. Strains of *E. coli*, *K. pneumoniae*, *Enterococci species*, *S. epidermidis* and *S. aureus* were isolated from hospitalized patients and well characterized according to standardized procedures in the Clinical Laboratory of the University of Patras. Before administration, 20 lots from each IVIG preparation were screened for antibodies against aforementioned bacterial strains. In general, titers of antibodies toward the bacterial surface antigens were lot-to-lot variable (CV ranging from 30 to 100%). Since administration of such lots could produce inconsistent results, the distribution of titers was studied for each bacterial isolate (see Fig. 1 for a characteristic histogram obtained for *E. coli*-specific antibodies). The distribution of titers for every bacterial pathogen followed an almost Gaussian distribution and therefore the majority of lots ($> 65\%$) contained titers in the range of $\bar{x} \pm \sigma$ ($\bar{x}$ being the mean value and σ the S.D.). Furthermore, the probability to obtain an unknown lot with antibody titer for a certain bacterial strain in the interval $\bar{x} \pm \sigma$ was higher than 65%. The administered lots were then selected so that the antibody titer for each strain was within this range, and consequently the lot-to-lot variability was minimized (Table 3). Nine out of 20 lots were selected for examining their effect in immunodeficient patients.

As shown in Table 3, high titers of antibodies specific to *E. coli* isolates tested were found in Sandoglobulin®, whereas such antibodies could not be detected in Gamimmune®. Sandoglobulin® also contained higher ($P < 0.001$) titers of antibodies towards a *S. epidermidis* strain. The con-

tent of specific antibodies to *Enterococci* was statistically ($P < 0.001$) higher in Gamimmune® as compared to Sandoglobulin®, whereas antibodies to *K. pneumoniae* and *S. aureus* in the two preparations were comparable. Notably, *S. aureus*-antibody titers were so high that they could not be compared even with the highest titers of the rest of the pathogens. This is ascribed to non-specific binding of Fc portion of IgG with protein A on the bacterial cell wall [9].

As shown in Table 4, levels of *E. coli*-specific antibodies obtained for the long-period Sandoglobulin®-treated patients, as well as 30 days after Sandoglobulin® infusion were significantly ($P < 0.001$) higher in patients' blood sera than those in healthy individuals. Following Gamimmune® infusion, however, the amounts of these antibodies were very low as compared to Sandoglobulin®-treated groups. This result can be well related with the composition of the IVIG preparations, since no specific antibodies to *E. coli* were identified in Gamimmune®. Amounts of antibodies to *K. pneumoniae* in immunodeficient patients were significantly ($P < 0.001$) higher than those in healthy individuals, suggesting that both IVIG preparations provided high amounts of such antibodies. Levels of *S. epidermidis*-specific antibodies in both long-period and short-term Sandoglobulin®-treated patients were significantly ($P < 0.001$) higher than those of healthy individuals. Following Gamimmune® infusion the amounts were very low as compared to Sandoglobulin®-treated patients, and comparable with levels of healthy individuals, since Gamimmune® contained lower amounts ($P < 0.05$) of the specific antibodies. Titers of antibodies against *Enterococci* spp. and *S. aureus* before and after each IVIG infusion were comparable to the control levels suggesting that despite differences between the two IVIG preparations in antibody titers, both provided sufficient amounts of pathogen-specific IgG.

4. Discussion

The advent of IVIG preparations signaled a new era in clinical immunology since their infusion is a safe, effective, and painless way of administering protective antibodies to immunodeficient patients. Retrospective studies on the outcome of prolonged treatment of primary immunodeficiencies (X-linked agammaglobulinaemia and hypogammaglobulinaemia) have demonstrated the effectiveness of IVIG in preventing severe infections [3,10]. In general, children who receive IVIG replacement therapy grow normally and have infection rates similar to those of non-immunodeficient children. Intravenous immunoglobulin exert anti-infective function through a variety of mechanisms. The most obvious mechanism is passive administration of antibodies specific to bacterial surface antigens which facilitate inhibition of microbial attachment, complement activation, and opsonization. There is also evidence that IVIGs contain neutralizing antibodies to infectious antigens, toxins, and bacterial superantigens. Furthermore, some of the immunomodulatory effects leading to downregulation of cytokine secretion, endothelial cell acti-

Table 4
Titers of specific antibodies toward bacterial pathogens (U ml^{-1}) in healthy individuals and patients' sera before and after IVIG administration

Bacteria	Healthy individuals	Long-period treated patients	After Sandoglobulin®	After Gamimmune®
E. coli	2.88 ± 0.71	4.89 ± 0.54*	4.61 ± 1.54*,**	2.91 ± 0.33
K. pneumoniae	2.05 ± 0.41	5.90 ± 1.10*	4.91 ± 0.70*	5.81 ± 1.20*
S. epidermidis	6.28 ± 1.41	10.8 ± 3.21*	11.50 ± 3.81*,**	6.87 ± 3.10
S. aureus	190.87 ± 67.34	222.43 ± 89.13	274.473 ± 62.45	312.73 ± 81.53
Enterococci spp.	5.06 ± 2.14	5.75 ± 2.31	5.51 ± 1.87	5.72 ± 1.32

* Statistically significant ($P \leq 0.001$) higher values comparing the IVIG-treated patients with healthy individuals.
** Statistically significant ($P \leq 0.001$) higher values comparing the Sandoglobulin®- and Gamimmune®-treated patients.

vation and lymphocyte function, which are hypothesized to improve autoimmune disease, may also prevent infection or minimize its consequences.

The standard way of treating patients with primary immunodeficiencies is to administer IVIG in low doses at frequent intervals to maintain the amount of immunoglobulins at levels able to protect patients against bacteremia. In this study, 10 male children with primary immunodeficiencies (common variable immunodeficiency and X-linked agammaglobulinaemia) were treated with IVIG. The dosage scheme applied to these patients resulted in serum IgG concentration higher than 7.0 g l^{-1}. This value has been proven critical since it is essential for the normal growth of children and the control of bacterial infections. Infusions were well-tolerated without any side-effects. Furthermore, the low pH of Gamimmune® did not affect the acid–base balance of the recipients and caused no discomfort.

In a recent study [6], differences of specific antibodies to Gram-negative bacterial strains between different IVIG preparations and between various lots from the same IVIG preparation were reported. These fine chemical differences triggered our concern to determine the levels of immunoglobulin classes and IgG subclasses, as well as pathogen-specific antibodies in patients. According to the World Health Organization (WHO), the distribution of IgG subclasses in IVIG preparations should reflect their occurrence in normal human serum (WHO Reference 67/97: $IgG_1 = 60\%$, $IgG_2 = 29.4\%$, $IgG_3 = 6.6\%$, $IgG_4 = 4.2\%$) [11]. The two IVIG preparations, Sandoglobulin® and Gamimmune®, exhibited significant differences in their composition. The concentrations of IgA, IgG_2 and IgG_4 were significantly higher in Sandoglobulin® and close to WHO standards, whereas IgG_1 concentration was higher in Gamimmune®. These differences may well be attributed to the different manufacturing procedures followed.

Screening of immunoglobulin classes and IgG subclasses in immunodeficient patients 30 days after IVIG administration revealed that both IVIG preparations affected their content in patients in different patterns. The most remarkable difference was noted for the concentration of IgG_4. Thus, IgG_4 levels in patients who received Gamimmune® were significantly lower than that observed in Sandoglobulin®-treated patients and this fact may well be attributed to the low levels of IgG_4 in the Gamimmune® preparation. The significance of low IgG_4 concentration is unclear at the time. In comparison with healthy individuals of the same median age and sex, all IVIG-treated immunodeficient patients had, in general, significantly lower concentrations of IgA and IgM following either Sandoglobulin® or Gamimmune® administration, due to the underlying defective immune response. IgG_1 was also significantly lower in patients than in the healthy individuals and was related with the low levels of total IgG and IgG_3. In general, low IgG_1 and IgG_3 subclasses suggest insufficient antibodies against proteins such as toxins produced by the diphtheria and tetanus bacteria, as well as antibodies against viral proteins.

Pathogen-specific IgG titers of 20 lots of each IVIG preparation were highly variable. Lot-to-lot variability in IVIG preparations has been described in earlier studies and, in general, has been blamed for inconsistent and conflicting results of clinical trials. In order to avoid discrepancies due to lot-to-lot variability of the amount of pathogen-specific IgG, following analysis and statistical evaluation of the titers for each pathogen tested, it was possible to select lots with reactivity in the interval $\bar{x} \pm \sigma$. Following this scheme, the lot-dependent effects on humoral status of patients were minimized and the results in patients can be assessed as preparation-specific. Although a limited number of clinically isolated bacterial strains were used, obtained results are strain-specific and very interesting conclusions can be drawn from this study.

Sandoglobulin® contained higher amounts of antibodies to *E. coli* and *S. epidermidis* than Gamimmune®, and its infusions resulted in maintenance of the levels in the recipients significantly higher than those in healthy individuals and Gamimmune®-treated patients. Correspondingly, estimated titers represented very high amounts of specific antibodies. Both IVIG preparations contained comparable amounts of specific

antibodies towards *K. pneumoniae*, which were sufficiently high since they kept recipients' levels twice as high as control levels. Gamimmune® contained higher amounts of *Enterococci* spp.-specific antibodies than Sandoglobulin® and both had comparable levels of antibodies to *S. aureus*. Titers of antibodies to *S. aureus* and *Enterococci* spp. in both preparations seem to represent a sufficient but not high enough amount of antibodies since the respective titers in patients' blood sera were close to those in healthy individuals. The sufficient pathogen-specific antibodies in patients were correlated with their serum IgG_2 (the concentration of which is similar to that of healthy individuals) and suggested satisfactory antibody response to polysaccharide bacterial antigens.

It was not possible to define a universal cut-off value with respect to the titer of specific antibodies to bacterial surface antigens which would allow immediate rejection of certain lots. However, we clearly showed a correlation of the composition of the infused IVIG preparations with the humoral immune status of treated patients. Screening of IVIG lots and selection of those having titers in the interval $\bar{x} \pm \sigma$, i.e. those with the highest probability to be received, seems to be helpful since administration of both IVIG preparations accomplished the maintenance of circulating pathogen-specific antibodies at levels comparable or significantly higher than those of the control group. High circulating levels in patients suggest successful prophylaxis against bacterial infections. The clinical profile of patients confirmed the results obtained by biochemical analysis. Therefore, although new ways of administering pooled human immunoglobulin emerge [12], intravenous infusion remains the commonest and safest clinical practice in the treatment of immunodeficient recipients.

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