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Intravenous immunoglobulin therapy for antibody deficiency

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SUMMARY

Twenty patients with antibody deficiency were treated at random with either intramuscular immune serum globulin (ISG) or intravenous modified immune serum globulin (M-ISG). Fourteen patients received of 259 M-ISG infusions during 242 months of treatment. Catastrophic vasomotor reactions were not observed. A single dose of 150 mg/kilo M-ISG increased serum IgG values a mean 248 mg%. Intravenous M-ISG therapy was effective in reducing the incidence of acute infections. Subjects receiving M-ISG developed 0·103 acute infections per month of treatment. Patients injected with ISG had 0·295 acute infections per month of treatment. Seven subjects had separate courses of both intravenous M-ISG and intramuscular ISG. Acute infections per month of treatment for M-ISG and ISG were 0·104 and 0·406, respectively.

INTRODUCTION

Many investigators have confirmed the beneficial effects of pooled, human immune serum globulin (ISG) in reducing infectious disease complications of agammaglobulinaemia (Barandum, Riva & Spengler, 1968; Domz & Dickson, 1957; Gitlin & Janeway, 1956; Janeway & Rosen, 1966). However, the intramuscular route employed limits its versatility and dosage. Intravenous administration is desirable and would solve many of these problems. Unfortunately, ISG infusions may induce catastrophic vasomotor reactions (Barandum *et al.*, 1962; Janeway, 1970; Merler *et al.*, 1967; WHO, 1966). The present report details our experience with a modified form of immune serum globulin (M-ISG) specifically prepared for intravenous usage.

MATERIALS AND METHODS

Patient. Twenty subjects with primary immunodeficiency (Table 1) were studied, amongst whom were nine subjects with X-linked agammaglobulinaemia and eleven with common variable agammaglobulinaemia. All patients satisfied criteria established by the World Health Organization for primary immunodeficiency (WHO, 1971).

Immune serum globulin preparation. Two forms of immune serum globulin, obtained from Cutter Laboratories, Inc., Berkeley, California, were used throughout this study. The intramuscular ISG reagent Gamastan® is commercially available and contains 16·5%± 1·5% gamma globulin dissolved in 0·3 M glycine. Intravenous M-ISG, patented by Cutter Laboratories, is produced by treating an aqueous solution of Cohn Fraction II with dithiothreitol to reduce and split selected inter-chain disulfide bonds. Alkylation with iodoacetamide is performed and excess reagents removed by cold ethanol precipitation and washing. The final product has little change of the IgG peak and approximately 90% sediments at 7S. The quantitative tetanus antibody precipitin assay indicates retention of 82% of specific activity. The anti-complement titre using sensitized sheep red cells drops from 1:512 to 1:1 for the final purified M-ISG. The plasma half-life determined by infusion of ^{125}I-labelled M-ISG into six recipients ranged from 15·5–28·7 days, with an average T 1/2 of 22·4 days. A 10% gamma globulin solution of this reagent was used.

Dosage and administration schedule. Patients were assigned to receive either intramuscular ISG or intravenous M-ISG on a random basis. The ISG group received the generally accepted schedule of 20 ml of ISG in two intramuscular injections,

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TABLE 1. Clinical and laboratory data of twenty primary immunodeficiency patients at the start of the study

Patients				Serum values of Ig (mg%)				
Number	Age	Sex	Diagnosis*	IgG	IgA	IgM	IgD	IgE
1	30	M	IX-LA	75	0	Trace‡	0	0
2	50	M	CVA	252	0	Trace	0	27
3	36	F	CVA	160	0	0	0	11
4	1	F	CVA	15	Trace	Trace	0	0
5	37	F	CVA	144	Trace	Trace	0	12
6	30	F	CVA	286	0	0	0	0
7	16	M	IX-LA	204	0	0	0	0
8	10	M	IX-LA	60	0	0	0	0
9	4	M	IX-LA	217	0	Trace	0	0
10	25	M	CVA	326	Trace	24	62	45
11	6	M	IX-LA	186	0	Trace	0	0
12	49	F	CVA	190	0	20	0	0
13	10	M	IX-LA	220	Trace	Trace	7	10
14	19	F	CVA	55	0	Trace	0	44
15	18	M	IX-LA	135	Trace	Trace	10	6
16	72	F	CVA	326	Trace	10	0	62
17	53	F	CVA	248	Trace	10	0	10
18	53	M	CVA	112	Trace	Trace	0	17
19	47	M	IX-LA	202	Trace	Trace	0	0
20	23	M	IX-LA	150	0	0	0	28

* IX-LA = Infantile X-linked agammaglobulinaemia; CVA = common variable agammaglobulinaemia.

† IgE values are international units/ml by radioimmunoassay. Normal values = 10–506 iu/ml; sensitivity of test 5 iu/ml.

‡ Trace = Faint reading too low to be quantified.

every 4 weeks. This optimum tolerated volume of repetitive intramuscular injections supplied 3·3 g of immune serum globulin. Several children received smaller volumes of ISG every 1–3 weeks because of restricted muscle mass.

M-ISG was infused at a dosage of 150 mg/kilo of body weight, every 4 weeks. The rate of infusion was controlled by an IVAC pump, with an average completion time of 4 hr. The 10% M-ISG solution was diluted for intravenous usage in two ways; an equal volume of M-ISG and 5% dextrose/water is currently employed. During the first year of this study the required volume of M-ISG was added to 200 ml of 5% dextrose/water solution. No therapeutic or symptomatic differences have been observed between these two diluting methods.

Patient evaluation. All patients completed a diary every 4 weeks covering work or school attendance, fever, antibiotics and the development of infection. Many subjects had chronic infections reflecting prior tissue damage. Chronic otitis media, sinusitis and bronchitis were frequently present and persisted. Accordingly, the development of acute infectious disease was closely monitored to determine the comparative therapeutic effects of ISG and M-ISG. An acute infection was diagnosed when a patient developed spiking fever greater than 100·4, with or without chills, clinical evidence of a purulent infection and positive cultures confirming the infection.

Laboratory studies. Serum levels of the five immunoglobulin classes were quantified prior to receiving the first therapy (Table 1). Several subjects were receiving monthly intramuscular ISG at the start of the study; one or more monthly therapies were withheld and immunoglobulin levels determined a minimum of 2 months after their last ISG administration. IgG, IgA, IgM, and IgD were quantified by the radial immunodiffusion method of Mancini, Carbonara & Heremans (1965). Commercial reference standards (Behringwerke) were used. IgE was determined using radioimmunoassay.

IgG values were determined prior to and directly after completing each M-ISG infusion. During the first 4 months of patient participation, IgG determinations were also performed 24 hr, 1 week, 2 and 3 weeks after receiving M-ISG. Six patients receiving intramuscular ISG had similar studies. IgG values were determined prior to, daily for 1 week and on the second, third and fourth week after receiving an intramuscular injection of ISG equal to 100 mg/kilo.

Prior to entering the study, periodically throughout the study and after the last M-ISG infusion, all sera were analysed for hepatitis surface antigen (HBsAg) by radioimmunoassay, and serum levels of bilirubin and glutamic-pyruvate transaminase (SGPT) were determined.

RESULTS

A total of 259 M-ISG infusions were administered to fourteen patients. Catastrophic vasomotor reactions were not encountered. Mild, transient effects (nausea, flush, fever, muscle cramps and headache) were common and related to the spread of infusion (Gerritz, Pirofsky & Nolte, 1976). They appeared to be vasomotor in origin, disappeared as the infusion was slowed and did not prevent administration of M-ISG A detailed review of these reactions will be presented elsewhere.

TABLE 2. Serum IgG levels prior to, directly after and 24 hr after receiving 150 mg/kilo M-ISG intravenously. Data obtained from nine subjects receiving a total of 100 infusions

		Serum values of Ig/mg%					
		Prior to M-ISG therapy		Post-M-ISG therapy		24 hr post-M-ISG therapy	
Patient number	Number of infusions	Mean	Range	Mean level	Mean rise	Mean level	Mean rise
2	15	220	176–318	423	203	425	205
3	6	117	98–136	354	237	293	176
5	15	334	185–445	651	317	512	178
7	15	190	127–316	443	253	352	162
8	7	292	156–371	523	231	393	101
11	15	136	90–199	371	235	332	196
12	8	280	190–329	477	197	417	137
13	11	238	176–292	564	326	459	221
14	8	173	55–277	403	230	345	172
Mean value		220	55–455	468	248	392	172

Serum levels of IgG in nine subjects receiving 100 infusions of M-ISG are presented in Table 2. A single infusion increased baseline values a mean 248 mg%. All subjects had increased serum IgG values above 350 mg% and seven out of nine above 400 mg%. After 24 hr, serum IgG was increased over pre-infusion levels by a mean 172 mg%. Accordingly, 30% of M-ISG had either been distributed into extra-vascular fluid, was tissue bound, or had been catabolized.

A similar study was performed with six subjects receiving 100 mg/kilo ISG intramuscularly. This standard therapy reflected the limitation of injectable volume imposed by muscle mass. In four out of six patients the maximum IgG serum level post-injection was found after 24–48 hr; in one patient it appeared at 1 week and in another 2 weeks. The mean maximum rise of serum IgG was 90 mg% above baseline values.

Sequential analysis of serum IgG levels revealed three patterns in subjects receiving M-ISG. Nine out of fourteen had a rapid rise of serum IgG, with a gradual reduction over a 4 week period and a return towards baseline values. In contrast, four patients had a slower progressive reduction of serum IgG levels over the 4 week period. IgG values were significantly elevated over baseline values prior to receiving their next M-ISG infusion. In one subject serum levels of IgG varied greatly, with rapid drops and elevations unrelated to M-ISG infusions.

Fourteen patients were treated with intravenous M-ISG for 7–28 months, with eight patients treated for more than 1 year (Table 3). A total of 259 infusions were given during a therapeutic period of 242 patient months. This group developed twenty-five acute infections (0·103 infections per month of treatment). These infections occurred in six subjects, with eight patients having 127 infection-free months. Patient No. 14 had the highest incidence of infectious disease, as well as cytomegalic disease and toxoplasmosis. Defects in T cell function were noted and progressively developing combined immunodeficiency was diagnosed. M-ISG therefore represented only a partial replacement therapy for this patient's immune defect. Without this patient, the infectious disease incidence for the intravenous M-ISG group was 0·067 acute infections per treatment month (Table 4).

TABLE 3. Incidence of acute infection in fourteen subjects receiving intravenous M-ISG

Patient number	Number of M-ISG infusions	Months of M-ISG therapy	Acute infections
1	7	7	1
2	29	26	6
3	8	7	0
5	29	28	4
7	29	27	0
8	7	7	0
10	23	23	0
11	27	27	3
12	25	22	0
13	23	22	0
14	25	19	10
16	9	9	0
17	10	10	0
18	8	8	1
Total	259	242	25
Mean	18·5	17·3	1·78
Range	7–29	7–28	0–10

Thirteen patients received 332 intramuscular ISG injections during 193 patient treatment months; fifty-seven acute infections were documented. Only two out of thirteen were free of infections for a period of 9 treatment months. Tables 4 and 5 list the complete data. Accordingly, patients receiving intramuscular ISG developed a mean of 0·295 acute infections per treatment month.

Separate courses of both intravenous M-ISG and intramuscular ISG were given to seven patients. These cross-over studies are not randomized and the data must be treated with caution. Subjects 8 and 16 left the M-ISG group to receive ISG because of discomfort or concern with intravenous M-ISG. Patients 1, 2 and 3 had frequent infectious complications while receiving intramuscular ISG. They were preferentially switched to the M-ISG intravenous route after infectious disease data had been accumulated. The rate of infectious complications was markedly reduced during the period when intravenous M-ISG was used. In spite of the limitation of not being random, the data are striking (Tables 4 & 6).

Statistical analysis of infection rates weighted for length of therapy was performed using the Mann–Whitney U (rank sum) test. The lower infection rate observed with M-ISG therapy was significant

TABLE 4. Data summary of infection rate in primary immunodeficiency patients receiving intravenous M-ISG and/or intramuscular ISG

	Intravenous M-ISG		Intramuscular ISG	Cross-over study	
	Entire group	Without No. 14	Entire group	Intravenous M-ISG	Intramuscular ISG
Number of patients	14	13	13	7	7
Number of therapies	259	234	332	112	167
Months of therapy	242	223	193	106	101
Numbers of acute infections	25	15	57	11	41
Acute infections/months therapy	0·103	0·067	0·295	0·104	0·406
Therapy months/acute infection	9·68	14·87	3·39	9·64	2·46

TABLE 5. Incidence of acute infection in thirteen subjects receiving intramuscular ISG injections

Patient number	Number of ISG injections	Months of ISG therapy	Acute infections
1	33	29	11
2	9	7	7
3	26	25	16
4	12	12	6
5	10	5	2
6	30	30	4
8	78	24	1
9	96	24	4
13	2	2	1
15	17	17	2
16	9	9	3
19	3	3	0
20	7	6	0
Total	332	193	57
Mean	25·5	14·9	4·38
Range	2–96	2–30	0–16

TABLE 6. Incidence of acute infections in seven subjects receiving separate courses of both intramuscular ISG and intravenous M-ISG

Patient number	Number of therapies		Months of therapy		Acute infections	
	M-ISG	ISG	M-ISG	ISG	M-ISG	ISG
1	7	33	7	29	1	11
2	29	9	26	7	6	7
3	8	26	7	25	0	16
5	29	10	28	5	4	2
8	7	78	7	24	0	1
13	23	2	22	2	0	1
16	9	9	9	9	0	3
Total	112	167	106	101	11	41
Mean	16·0	23·9	15·1	14·4	1·57	5·86
Range	7–29	2–78	7–28	2–29	0–6	1–16

($P<0·05$), if patient 14 with combined T and B cell defects is excluded, the difference is significant ($P<0·01$). In the study involving patients receiving separate courses of both ISG and M-ISG, the M-ISG lower infection rate was significant ($P<0·05$).

Hepatitis did not result from the intravenous administration of M-ISG. HBsAg testing was negative and serum levels of bilirubin and glutamic-pyruvate transaminase (SGPT) remained normal throughout the study.

DISCUSSION

Previous studies have indicated that ISG significantly reduces infectious disease complications in patients with agammaglobulinaemia (Barandun *et al.*, 1968; Domz & Dickson, 1957; Gitlin & Janeway, 1956; Janeway & Rosen, 1966). Intramuscular therapy is employed because intravenous use induces severe vasomotor reactions (Barandun *et al.*, 1962; Janeway, 1970; Merler *et al.*, 1967; WHO, 1966). Anti-complementary globulin aggregates apparently create such reactions by releasing vasoactive substances (Janeway, 1970; WHO, 1966; Gerritz *et al.*, 1976; Ishizaka *et al.*, 1967; Schiff, Sutherland & Lane, 1968).

The intramuscular route imposes severe restrictions on the effective clinical use of ISG (Hitzig & Muntener, 1975). Several days are required to reach maximum blood levels of antibody, thereby limiting its use to routine prophylaxis (Smith *et al.*, 1972). A depot effect occurs which may modify the functional capability of antibody. Increased catabolism and distribution into the extravascular compartment reduces blood levels of IgG. In addition, the volume of injected ISG is limited by the available muscle mass of the patient. This can lead to either an inadequate dosage or more frequent injections. Discomfort resulting from frequent intramuscular injections of large volumes may discourage both patient and physician, interfering with effective therapy.

Several attempts have been made to produce an intravenous form of ISG. Acid and pepsin treatment of ISG have been employed to reduce vasomotor reactions (Janeway, 1970; WHO, 1966; Ishizaka *et al.*, 1967; Schultz & Schwick, 1962). Such treatments create IgG with a short half-life, thereby limiting clinical usefulness (Barandun *et al.*, 1968; Koblet, Barandun & Diggelmann, 1967; Wells & Penny, 1969). Deaggregation by the human enzyme plasmin has been similarly used (Sgouris, 1967). Treatment sufficient to reduce vasomotor effects, however, also degrades the globulin molecule, reducing its half-life (Janeway *et al.*, 1968; Painter, Walcroft & Weber, 1965). Hainski *et al.* (1971) employed a polymeric fractionation agent to purify Cohn Fraction III; this preparation transmits hepatitis and cannot be recommended.

The present study has evaluated a new preparation of modified immune globulin, M-ISG, specifically produced for intravenous administration. Mild disulfide reduction is employed to reduce globulin aggregation and essentially eliminate anti-complement activity. The resulting globulin solution maintains antibody activity and has a satisfactory half-life for long-term therapy. Catastrophic vasomotor reactions did not occur during 259 infusions. Mild vasomotor reactions were frequently encountered. These were transient, related to the rate of infusion and did not interfere with its clinical application (Gerritz *et al.*, 1976).

Sequential analysis demonstrated that monthly infusions of 150 mg/kilo of M-ISG were effective in elevating IgG serum values. Blood ranges of IgG were increased an average of 248 mg% directly following infusion, with a mean IgG blood level of 468 mg%. In contrast, maximum serum IgG elevation following monthly injections of 100 mg/kilo ISG was only 90 mg%. The intravenous route using M-ISG accordingly offers a highly desirable method of elevating serum IgG into therapeutic ranges.

The current study indicates that M-ISG therapy is effective. Twenty patients were given, at random either intravenous M-ISG or intramuscular ISG. Acute infections occurred at a mean monthly rate of 0·103 and 0·295, respectively, for these two agammaglobulinaemic groups. Seven patients received separate courses of both M-ISG and ISG. This cross-over group was selective and not picked at random. Patients receiving intravenous M-ISG had an acute infection monthly rate of 0·104; the same patients receiving intramuscular ISG developed a mean of 0·406 acute infections per month.

It should be emphasised that subjects in the M-ISG group received substantially more IgG than the ISG study patients. The intravenous route permitted an ideal, calculated quantity of IgG to be infused, establishing a therapeutic state. Administering similar quantities of IgG to the ISG population would have required either more frequent intramuscular injections and/or painfully large volumes. Patient acceptance of the intravenous M-ISG therapy was excellent. Subjects were aware of fewer infectious complications, able to reduce or discontinue antibiotic therapy, lost less time from school or work, and noted a progressive feeling of well-being.

Intravenous M-ISG is practical for the long-term therapy of agammaglobulinaemia. An additional use deserves evaluation. In contrast to intramuscular ISG, intravenous M-ISG produces immediate high blood levels of effective antibody and may be useful in the treatment of acute infectious diseases including viremic episodes. Further studies are required to examine the potential benefits of M-ISG in iatrogenic or disease-induced immunodeficiency complicated by infection.

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