INTRODUCTION

Requirements for safety and quality of intravenous immunoglobulin G preparations

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Abstract

The increasing use of intravenous immunoglobulin-G preparations (IVIg) calls for evaluation of the requirements for quality and freedom from infection.

The WHO Requirements dating from 1982 have been surpassed by the majority of IVIg preparations. Three methods are used for ensuring safety from infection: the restrictive choice of donor, the precise introductory screening of every individual blood or plasma donation, and the additional active virus inactivation procedures that surpass the virus elimination of, for example, the Cohn-fractionation method. Although it is highly improbable that IVIg can transfer infections, this possibility cannot be completely ruled out even when all the GMP (good manufacturing practice) rules are observed most carefully. Increased safety from infection would be provided if blood and plasma donations were obtained nationally from unpaid voluntary donors and if an additional active viral inactivation procedure was introduced.

IVIg has assumed an important role in prophylaxis and therapy of bacterial and viral infectious diseases for a considerable period, as well as in the treatment of primary and secondary antibody deficiency syndromes.

Various new indications for IVIg have appeared during the last few years and have been described and reviewed many times in previous reports^{1-4,15} and in this supplement. IVIg therapy has gained increased importance in the treatment of HIV infections,² and particularly in autoimmune diseases.³ Although it is highly unlikely that IVIg preparations have caused transferred infection, and even though a distinct improvement in the safety of these preparations has been achieved during the last 20 years, it is still necessary to define quality and safety standards.

QUALITY REQUIREMENTS OF IVIg PREPARATIONS

The WHO requirements for intravenous immunoglobulin preparations stem from 1982 and lay down minimum standards.

IVIg should be extracted from a pool of at least 1000 individual donors. The IgG preparations should contain at least 90% intact IgG and as small an amount of IgA concentration as possible, as well as being free from fragments and aggregates.

IVIg should be modified biochemically as little as possible and should possess opsonising and complement fixing characteristics as well as other natural biological characteristics.

All IgG subclasses should be present, where possible, in similar distribution as in normal human plasma. The immunoglobulins should meet WHO standards and be free from prekallikrein activator, kinins, plasmins, accumulating preservatives (stabilisers) and other damaging contaminants as far as possible.⁶

These requirements are clearly surpassed today by the majority of manufacturers. Acute intravenous side effects of IVIg preparations are therefore not a relevant problem today.²

SAFETY REQUIREMENTS FROM INFECTION THROUGH IVIg

A risk of infection from human plasma preparations can never be completely ruled out. The point to be discussed therefore is which methods are at our disposal.

Both the national health authorities and the WHO have issued corresponding guidelines that guarantee to the greatest possible extent the safety of blood products.⁷ These mainly cover the following points:

1) The best possible quality of the initial plasma (blood or blood plasma donations)

A careful selection of donors is necessary. The guidelines issued by the EC on 14 June 1989 emphasise the importance of voluntary, unpaid blood donation.⁸ The careful medical examination of every individual donor as well as appropriate questionnaires for anonymously eliminating any "high-risk" donors constitute further safeguards.

Table 1 IgG subclasses in WHO reference plasma

Subclasses	WHO reference plasma 100% IgG
IgG ₁	60,0%
IgG ₂	29,4%
IgG ₃	6,5%
IgG ₄	4,1%

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Table 2 Possible pathogens in blood and blood derivatives

Viruses	Lipoprotein-envelope	Blood	Blood-derivatives
HBV	+	+	+
HCV	+	+	+
HNANB* (HCV)	+	+	+
HIV	+	+	+
HTLV-1	+	+	—
CMV	+	+	—
EBV	+	+	—
Parvovirus B19	—	—	+
HAV	—	+	—
Jakob-Creutzfeldt	—	—	—

*Hepatitis non A—non B.

3) Hepatitis Delta Virus (HDV)

4) Human Parvovirus

5) Hepatitis C Virus (HCV).

Various virus-inactivation procedures are currently employed for the purpose of optimising the safety of plasma products.

VIRUS INACTIVATION PROCEDURE WITH IVIg

Virus removal by Cohn-fractionation, with particular reference to HIV, has already been comprehensively described.¹²

The occurrence of HCV infection after IVIg treatment led to safety improvement in the manufacturing procedures.

Virus removal is achieved by the following measures which are employed in addition to the cold ethanol fractionation:

- 1) Reduction in the pH values to 4.25 with subsequent incubation of the finished preparation at pH 4.25 and 25°C for 24 days
- 2) Beta-propiolactone treatment
- 3) Pasteurisation, 10 hours at 60°C
- 4) Oxidative S-sulphonation
- 5) Hydrolases bound to fixed phases.
- 6) Solvent/detergent treatment with TNBP (tri-(n-butyl)-phosphate)/Na-cholat resp. Tween 80 and sodium cholate.

Experience has shown that the chemical or physical virus inactivation procedures employed in the manufacturing process impair the development of the IVIg preparations. As a rule, additional auxiliary substances and stabilisers must then be used to prevent changes in the structure and functional capacity of the individual native antibody molecule and also to prevent a change in the composition of subclasses IgG₁₋₄.

VALIDATION

Various studies are performed to validate the viral inactivation potential of a manufacturing process. Such studies consist of testing every single manufacturing step for its viral inactivation potential with regard to a specific virus.

As a rule, a defined quantity of virus is added to the starting material. Test samples are run in duplicates which are treated identically. The respective samples are examined for their viral content at the end of each selected manufacturing step. The value found is then compared with the quantity of virus which was added initially to the starting material.

Model viruses, as well as host-specific

Table 3 Example of a reduction through Cohn-fractionation

Viruses	Reduction (log ₁₀)
HIV-1 (HIV-2)	>15.6
Bovine viral diarrhoea virus (HCV)	6.6
Pseudorabies virus (HBV)	14.4
Simian virus 40	5.1

viruses, are selected for the studies concerned.

1) Bovine viral diarrhoea virus

Model virus for HCV, enveloped RNA virus which can be slightly inactivated by heat, is sensitive to organic solvents, acid-labile, base-proof.

2) Pseudorabies virus

Model virus for HBV, enveloped DNA virus, relatively thermolabile, sensitive to organic (lipophilic) solvents and extreme pH values.

3) Simian virus 40

Model virus for non-enveloped DNA viruses, very stable virus, resistant to ether, 2% phenol and 50% ethanol.

4) HIV-1

Retrovirus, serves at the same time as a model virus for HIV-2.

The data for the elimination rates are expressed logarithmically. The reduction factor is calculated using the following formula:

$$\frac{\text{Total amount of virus in the starting material}}{\text{Total amount of virus in the test sample}}$$

Table 3 shows an example of typical elimination rates using Cohn-fractionation as the sum of the individual manufacturing steps (overall reduction factor).

While the elimination rates of the model virus for HIV and HBV appear to be sufficient, the reduction in HCV and non-enveloped viruses is distinctly lower. It has already been mentioned that due to the unlikely risk of viral infection from IVIg, these preparations have been given little attention. From a current standpoint, however, the introduction of a further manufacturing step for the purpose of viral inactivation—in addition to the viral elimination potential of the Cohn-fractionation procedure—should be made a requirement for the manufacture of IVIg preparations.

It has also been demonstrated that commercial and paid blood donations increase the risk of infection with hepatitis pathogens. It has been shown that infection with hepatitis considerably increases in relation to the number of paid blood donors.¹³ The EC commission guidelines therefore recommend using unpaid voluntary donors.⁸ This recommendation also provides a better guarantee of a balanced antibody spectrum of the IVIg preparations for a given population.¹⁴

Conclusion

The 1982 WHO requirements form the minimum basis world wide for the quality and

safety of IVIg. Because the range of application of IVIg has expanded considerably in the last 10 years, there is a fundamental requirement to introduce further active virus inactivation procedures which supplements the virus elimination achieved with, for example, the Cohn-Oncley-fractionation method.

This applies above all for IVIg preparations which are not obtained from blood and plasma from unpaid voluntary donors in the framework of a national self-supply, since such sources are characterised by a higher risk of infection—particularly when they are collected in social fringe groups of population centres.¹³

Because the various active virus inactivation procedures exhibit different specificity of action for certain viruses, an IVIg preparation would be ideal—according to current knowledge—if it were obtained nationally from voluntary, unpaid blood or blood plasma donations and subjected to two active virus elimination procedures with different mechanisms of action.

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