Pharmacokinetic Studies with Recombinant Cytokines
Scientific Issues and Practical Considerations

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Summary

Advances in molecular biology and recombinant DNA technology have led to the development of cytokines as therapeutic agents for a variety of disease states. The pharmacokinetic analysis of cytokines involves the understanding of analytical methods capable of detecting these agents in biological fluids and recognition of several factors which may have an impact on the cytokine concentration-time curves.

Enzyme-linked immunosorbent assays (ELISA) have become the most common method of detection and commercial kits are available for a wide variety of cytokines. Monoclonal antibody products are sensitive, have minimal cross-reactivity and are relatively inexpensive when compared with high performance liquid chromatography (HPLC). However, the primary limitation of these assays is their inability to measure biologically active protein. Conversely, bioassays
do measure a biological event (i.e. proliferation or cytotoxicity) but are generally not used for cytokine analysis because of their high cost, long assay completion time, lack of specificity, poor sensitivity and influence of environmental conditions on the outcome.

The pharmacokinetic profile of recombinant cytokines is influenced by a number of variables: endogenous production, circulating soluble receptors and cell-associated receptors, immunocompetence and antibody production against the cytokine all may influence the disposition of the agent. Thus, pharmacokinetic modelling of cytokines may involve complex models capable of characterising these nonlinear processes and resulting effects.

The route of administration is an important variable since cytokines administered by subcutaneous injection may be partially metabolised by proteases present in the subcutaneous tissue. Other methods to simplify cytokine delivery are being actively investigated and include formulations for inhalation, topical and oral administration. A variety of cytokines (including interferon-α, interleukin-6 and tumour necrosis factor) are capable of inhibiting cytochrome P450 hepatic enzymes and, therefore, possess the potential to cause drug-cytokine interactions. Inhibition has been demonstrated in several in vitro systems and animal models, although clinical data are currently limited.

An increased understanding of the many factors which can alter the analysis and pharmacokinetics of cytokines is essential to the design of optimal dosage regimens.

Scientific advances in immunology and recombinant DNA technology have led to the development of endogenous cytokines as therapeutic modalities. These glycoprotein molecules have multiple biological activities which are currently being actively evaluated as drugs for the treatment of a variety of diseases, including cancer, AIDS, sepsis, hepatitis and multiple sclerosis (table I).

The study of the pharmacokinetics of recombinant biological products involves the understanding of multiple factors which may influence their disposition, such as endogenous production, receptor binding effects and time-dependent pharmacokinetics. Other factors such as formulation, route of administration, distribution variability and time of infusion also require consideration as they may dramatically alter the pharmacokinetic profile.

Data analysis of biological proteins also presents a challenge. The common methods of pharmacokinetic analysis often do not adequately describe the area under the concentration-time curve (AUC) profile of biological products. Most agents do not conform to standard compartmental models with first order elimination. In addition, it is important to note that data which may appear to be outliers or errant points may indeed reflect the true pharmacokinetic profile of the agent under study. Thus, a combination of factors play a role in the accurate description of biological drug disposition. In this review we focus on understanding the complexity of characterising pharmacokinetic parameters associated with the exogenous administration of recombinant cytokines.

1. Analytical Methods for Quantification

Assay specificity and sensitivity are the primary obstacles involved in the quantification of conventional pharmaceuticals. These obstacles are amplified when working with cytokines because of endogenous protein production and the fact that the methods commonly employed in pharmaceutical analyses [i.e. high performance liquid chromatography (HPLC) and gas chromatography] may not be applicable for assay of these proteins; thus, alternative methods are necessary [i.e. enzyme-linked immunosorbent assays (ELISA), radioimmunoassays and bioassays].