Microdosing of the human protein hRESCAP in healthy volunteers to predict its clinical pharmacokinetics

For the development of New Biological Entities (NBEs) such as human protein therapeutics, considerable costs and time are spent on preclinical animal studies, which are often not predictive for the situation in humans. Microdosing, combining with the ultrasensitive analytical technique of Accelerator Mass Spectrometry (AMS), provides a safe and reliable approach to obtain first-in-human pharmacokinetic data, after limited animal testing. We investigated whether microdosing can be used as a safe starting dose tool to study pharmacokinetics and safety of human recombinant Placental Alkaline Phosphatase (hRESCAP), a protein currently under investigation for clinical applications.

Methods: hRESCAP was produced and [14C]-labelled under GMP-compliance. hRESCAP and [14C]-hRESCAP, or placebo (n=3 per group) were i.v. administered to healthy volunteers in a single ascending dose design. For the development of New Biological Entities (NBEs) such as human protein therapeutics, microdosing of 14C-labelled biotherapeutics preceded by a rationally designed preclinical package can substantially reduce the time and costs of drug development, as well as the number of laboratory animals used. The pharmacokinetics across dose groups and individuals was observed, indicating dose linearity from microdose to therapeutic doses. Pharmacokinetic analysis of [14C]-hRESCAP at various doses indicated a dose linear behavior of the radiolabel in healthy volunteers. At the highest dose, total plasma levels were ~3-fold above endogenous AP levels prior to ~4.5 days after administration (not shown). Pharmacokinetic analysis of [14C]-hRESCAP at various doses indicated a dose linear behavior of the protein (Figure 2B). The terminal half life of [14C]-hRESCAP was calculated to be 4.8 days. Furthermore, data from the microdose could be used to accurately predict levels of the enzyme at higher doses (Figure 2C).

RESULTS

A microdose of [14C]-hRESCAP could be used as a safe starting dose for a first-in-human study, and to predict the pharmacokinetics of hRESCAP at therapeutic doses. hRESCAP was well tolerated in healthy subjects up to 5300 µg, and had a favorable half-life in humans. Microdosing of 14C-labelled biotherapeutics preceded by a rationally designed preclinical package can substantially reduce the time and costs of drug development.

CONCLUSIONS

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ABSTRACT

hRESCAP was extensively characterized by in vitro methods, and ethical approval for a first-in-human phase 0 / phase 1 study was obtained. A microdose (53 µg, 0.5 nmol) of [14C]-hRESCAP was intravenously (i.v.) administered as a safe starting dose to healthy volunteers (n=3), for safety and pharmacokinetics assessment. Subsequent doses of 414, 1240 and 5300 µg (including 53 µg [14C]-hRESCAP), or placebo (n=4 per group) were i.v. administered according to a single ascending design. [14C]-hRESCAP plasma levels were determined with AMS and total hRESCAP levels with an enzymatic assay. Plasma concentration-time data were analyzed by nonlinear mixed-effects modeling.

Figure 1. Clinical Phase 0 / Phase 1 study with [14C]-hRESCAP. A) Study design. The set up of the study is depicted in the scheme (left panel). In short, a microdose of [14C]-hRESCAP alone or supplemented with hRESCAP was administered to healthy male volunteers and blood was collected at various time points. After collection, [14C]-hRESCAP and [14C]-hRESCAP (including stability, (radio)chemical purity, enzymatic activity, SDS-PAGE, mass spectrometry of the glycosylated and deglycosylated product, and determination of protein aggregation by SEC-UV) and limited animal testing (2-week repeated dose toxicity test in human Alkaline Phosphatase tolerized mice), approval from the Dutch Medical Ethical committee for a clinical Phase 0 / Phase 1 first-in-human study with [14C]-hRESCAP and hRESCAP was obtained.

pharmacokinetics across dose groups and individuals was observed, indicating dose linearity from microdose to therapeutic doses. Pharmacokinetic analysis of [14C]-hRESCAP at various doses indicated a dose linear behavior of the protein (Figure 2B). The terminal half life of [14C]-hRESCAP was calculated to be 4.8 days. Furthermore, data from the microdose could be used to accurately predict levels of the enzyme at higher doses (Figure 2C).