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ORIGINAL ARTICLE

Modelling inflammatory biomarker dynamics in a human lipopolysaccharide (LPS) challenge study using delay differential equations

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Clinical studies in healthy volunteers challenged with lipopolysaccharide (LPS), a constituent of the cell wall of Gram-negative bacteria, represent a key model to characterize the Toll-like receptor 4 (TLR4)-mediated inflammatory response. Here, we developed a mathematical modelling framework to quantitatively characterize the dynamics and inter-individual variability of multiple inflammatory biomarkers in healthy volunteer LPS challenge studies. Data from previously reported LPS challenge studies were used, which included individual-level time-course data for tumour necrosis factor α (TNF-α), interleukin 6 (IL-6), interleukin 8 (IL-8) and C-reactive protein (CRP). A one-compartment model with first-order elimination was used to capture the LPS kinetics. The relationships between LPS and inflammatory markers was characterized using indirect response (IDR) models. Delay differential equations were applied to quantify the delays in biomarker response profiles. For LPS kinetics, our estimates of clearance and volume of distribution were 35.7 L h^{-1} and 6.35 L, respectively. Our model adequately captured the dynamics of multiple inflammatory biomarkers. The time delay for the secretion of TNF-α, IL-6 and IL-8 were estimated to be 0.924, 1.46 and 1.48 h, respectively. A second IDR model was used to describe the induced changes of CRP in relation to IL-6, with a delayed time of 4.2 h. The quantitative models developed in this study can be used to inform design of clinical LPS challenge studies and may help to translate preclinical LPS challenge studies to humans.

KEYWORDS
biomarkers, endotoxaemia, inflammation, LPS, modelling

There is no principal investigator because this is a secondary analysis of a previously published clinical study. Co-author M. Moerland, however, coordinated the clinical studies used in this analysis.

1 | INTRODUCTION

Systemic inflammation plays a pivotal role in a multitude of conditions, including sepsis, trauma, major surgery and burns.[1] The associated inflammatory response in systemic inflammation is a complex dynamic response of various cytokines,[2] including **interleukin (IL)-6**, **IL-8** and **tumour necrosis factor α (TNF-α)**, and acute phase proteins, including C-reactive protein (CRP).[3]

Inflammatory markers in systemic inflammation are of potential utility to guide treatment decision-making and optimization of antibiotic treatments,[4] as they could indicate if a treatment is able to modulate the response to an infectious challenge. Currently, CRP is commonly applied to inform such decision-making.[3] However, the relatively long half-life[5] of CRP has limited its utility to make timely decisions on treatment adjustments, which is crucial for treatment of severe infections and sepsis.

Several of the inflammatory cytokines have been investigated as targets for anti-sepsis treatments.[2] While agents inhibiting TNF-α or IL-1β showed positive results in preclinical studies, they did not lead to improvements in therapeutic outcomes in septic patients.[6] On one hand, the failure to find effective drugs to modulate the systemic inflammatory response in sepsis may be explained by the highly heterogeneous character of the underlying pathophysiology. In addition, translational gaps for systemic inflammation may be a cause of this problem. Enhancing our understanding of quantitative differences in the dynamics of the inflammatory response between preclinical animal models and patients may help to address this gap.[7,8] However, the characterization of host response in critically ill patients is challenging, partly due to the large underlying variation in disease, patient and treatment-related factors.

Human healthy volunteer models of the inflammatory response to infectious challenges are of relevance as an intermediate disease model to close the translational gap towards patients, and to further characterize potential biomarkers to support treatment optimization. Human healthy volunteer endotoxaemia models allow the induction of an inflammatory response in a controlled setting. In such studies, healthy volunteers are typically administered intravenous **lipopolysaccharide (LPS)**, which activates **Toll-like receptor 4 (TLR4)**, leading to an increased production of inflammatory markers including various cytokines and CRP.[9]

Quantitative characterization of the dynamics of inflammatory cytokines can enhance our understanding of the inter-species differences during drug development and guide the application of these biomarkers for treatment response monitoring. To this end, a number of mathematical models have been developed to quantitatively describe the time-course of inflammatory markers in animals after LPS administration.[10,11] However, such models have not been used for translational predictions and analysis of human LPS challenge studies. Developing a model of human biomarker dynamics is therefore an essential step towards the construction of a quantitative translational framework describing between-species differences in inflammatory response, and informing the study design of novel human LPS challenge studies. In this study, we aimed to develop quantitative models of inflammatory marker dynamics, including TNF-α, IL-8, IL-6 and CRP, in response to an LPS challenge in healthy volunteers.

What is already known about this subject

- Lipopolysaccharide (LPS) challenge studies in healthy volunteers are a relevant clinical model to study the Toll-like receptor 4 (TLR4)-mediated inflammatory response in humans.

What this study adds

- We quantitatively describe the dynamics of multiple inflammatory proteins in response to LPS exposure.
- The developed mathematical models can serve as a translational tool in drug research of inflammatory biomarkers and investigational drug treatments targeting the inflammatory response.

2 | METHODS

2.1 | Data

Data derived from three previously conducted LPS challenge studies in healthy volunteers were used for model development (Table 1). Briefly, Study A characterized the mean LPS concentration–time profile in six healthy volunteers after a single low dose (2 ng kg^{-1} body weight) administered as an intravenous bolus injection.[12] Studies B and C characterized the dynamics of multiple inflammatory markers after LPS administration. Study B[13] consisted of three cohorts of eight subjects (LPS:placebo = 6:2) who received a single ascending low dose of LPS (0.5, 1.0 or 2.0 ng kg^{-1}). Study C[14] involved four volunteers following a single LPS dose of 2.0 ng kg^{-1}. A series of blood samples was drawn longitudinally to measure the cytokine (TNF-α, IL-6, IL-8) concentrations up to 48 hours post dose and 96 hours post dose for CRP. In total, 327 TNF-α samples, 288 IL-8 samples, 327 IL-6 samples and 211 CRP samples from 28 healthy volunteers were used for model development. Studies B and C used different CRP assays, with different sensitivity, yielding the lowest recorded CRP concentrations to be 0.04 and 3.0 mg L^{-1}, respectively. No information about the limit of quantification (LOQ) was available for either of the studies, thus the lowest recorded values were chosen as a proxy for LOQs.

TABLE 1 Overview of available samples from published studies

Study	Number of subjects	LPS dose (ng kg^{-1})	Measured markers	Sampling strategies	Number of available samples (BLOQ)
A[12]	6	2.0	LPS	0, 15. 30, 45, and 60 minutes and thereafter at half-hourly intervals for a total period of 6 hours[a]	6 mean values (50%)
B[13]	24	0.5, 1.0, 2.0 and placebo	TNF-α	Before and 6, 12, 24, 48 and 72 hours[a] after dose	288 (0)
			IL-8		288 (0)
			IL-6		288 (6.25%)
			CRP		144 (0)
C[14]	4	2.0	TNF-α	Before and 6, 12, 24 hours and thereafter at 24-hourly intervals up to 8 weeks[a] after dose	43 (0)
			IL-6		43 (0)
			CRP		67 (65.67%)

Abbreviations: LPS: lipopolysaccharides; TNF-α: tumour necrosis factor α; IL-8: interleukin 8; IL-6: interleukin 6; CRP: C-reactive protein; BLOQ: below the limit of quantification.

[a]LPS was undetectable after 1 hour, all three cytokines were undetectable after 48 hours, and CRP was undetectable after 96 hours.

2.2 | Model implementation

We first developed a model describing the kinetics of LPS, based on Study A. We then developed models to characterize the dynamics of TNF-α, IL-8, IL-6 and CRP, as available from Studies B and C. Delay differential equations (DDEs) were implemented to capture the time delay[15] between exposure of LPS and the release of inflammatory cytokines, and between IL-6 exposure and CRP release. All models were fitted using the stochastic approximation expectation maximization (SAEM) and importance sampling (IMP) methods with DDE solver implemented in NONMEM version 7.5. For samples with below the limit of quantification (BLOQ), the M3 method[16] was used. (The model code is provided in the Supporting Information).

2.3 | LPS kinetics

Standard one- and two-compartment models with either linear or nonlinear elimination were evaluated to describe typical LPS kinetics. No inter-individual variability (IIV) was estimated due to the lack of sufficient individual data. Additive, proportional and combined residual error models were tested to capture the residual unexplained variability.

2.4 | Inflammation marker dynamics

2.4.1 | Cytokine dynamics

The relationship between plasma concentrations of LPS (C_{LPS}) and cytokines ($C_{cytokine}$) was modelled using indirect response (IDR) models with a delayed concentration-dependent stimulation by LPS on the production of cytokines. The IDR models described the change of cytokine concentrations, consisting of a zero-order secretion rate (k_{in}), a first-order degradation rate constant (k_{out}) and a baseline given by the ratio k_{in}/k_{out} at steady state. Nonlinear secretion rates were also investigated. The stimulatory effect of LPS concentration (S_{LPS}) concentration was tested using linear, exponential, power, E_{max} and Hill functions. DDEs were implemented to address the observed effect delay between LPS concentration and LPS stimulus on cytokine secretion. The cytokine dynamics were described according to Equation (1), where effect delay was captured using a delay time factor (τ).

$$\frac{d(C_{cytokine})}{dt} = k_{in} \cdot (1 + S_{LPS} \cdot C_{LPS}(t-\tau)) - k_{out} \cdot C_{cytokine}, \quad (1)$$

where $C_{cytokine}$ represents the concentration of cytokine TNF-α, IL-8 and IL-6, respectively, in their individual models.

2.4.2 | C-reactive protein dynamics

CRP was assumed to be exclusively stimulated by IL-6 based on a previous in vitro study in hepatoma cell lines.[17] Therefore the above indirect response model was adapted for CRP (Equation 2).

$$\frac{d(C_{CRP})}{dt} = k_{in} \cdot (1 + S_{IL-6} \cdot C_{IL-6}(t-\tau)) - k_{out} \cdot C_{CRP}. \quad (2)$$

Profiles of the inflammatory markers in healthy volunteers ($n = 6$) that received placebo injections (Figure S1) showed high within-patient variability in the concentrations of IL-6 and CRP without LPS challenge. Relatively high variability and a declining trend were observed in the late phase of IL-6 and CRP upon LPS challenge (Figure S2), thus the assumption of stable state was abandoned and individual baseline values were estimated for both of these markers instead of being derived by the ratio k_{in}/k_{out}. In addition, a time-dependent secretion rate was tested to describe the potential decline of IL-6 secretion over time with an exponential decay factor $k_{decline}$ (Equation 3).

$$k_{in} = k_{in}0 \cdot e^{(-k_{decline} \cdot t)}, \quad (3)$$

where $k_{in}0$ is the IL-6 secretion rate immediately following LPS injection.

2.4.3 | Inter-individual and residual unexplained variability

Inter-individual variability (IIV) was tested on all parameters in the inflammation marker models for the quantification of variability in the study population. The random effect η was assumed to follow a normal distribution with a mean of 0 and a variance of ω^2 (Equation 4).

$$P_i = TVP \cdot \exp(\eta_i) \quad \eta \sim N(0, \omega^2). \quad (4)$$

Here, P_i is the parameter value for an individual i, TVP is the typical population value of the parameter, and η_i are inter-individual random effects for an individual i and the parameter P.

Correlation between random effects was investigated for all parameters. Additive, proportional and combined residual error models were tested to capture the residual unexplained variability.

2.4.4 | Model evaluation

Model evaluation was based on numerical and graphical diagnostics. Nested models were compared using objective function value (OFV), where a ΔOFV > 3.84 in case of one degree of freedom (i.e., $P < .05$) was considered significant. The precision of the parameter estimates was evaluated using relative standard error of the estimate (RSE%). Graphical diagnostics included basic goodness-of-fit (GOF) plots, and prediction corrected visual predictive checks (pcVPCs) based on 1000 simulations obtained with software Perl-speaks-NONMEM (PsN) version 4.9.0.[18]

2.5 | Nomenclature of targets and ligands

Key protein targets and ligands in this article are hyperlinked to corresponding entries in https://www.guidetopharmacology.org, and are permanently archived in the Concise Guide to PHARMACOLOGY 2021/22.[19]

3 | RESULTS

3.1 | LPS kinetics

The observed mean LPS time-course profile was adequately described using a one-compartment model with first-order elimination (Figure S3). Clearance (CL) and volume of distribution (V) were estimated to be 35.7 L h^{-1} and 6.35 L, respectively, with a proportional residual error of 32.5%. CL and V were subsequently fixed to generate a typical LPS concentration–time profile used in the modelling of the inflammatory markers.

3.2 | Cytokine dynamics

A modelling framework (Figure 1), consisting of a series of IDR models developed for each inflammatory marker, was developed which could adequately capture the inflammatory marker time-course profiles (Figure 2, Figure S4A–D). The relationships between LPS concentration and secretion of TNF-α, IL-6 and IL-8 were described by IDR models with linear stimulations on the secretion of cytokines, in combination with DDEs to account for the delay. Parameter estimates for each model (Table 2) showed adequate precision.

For TNF-α and IL-8, the baseline values were assumed to be represented by the ratio of secretion rate constant (k_{in}) and the degradation rate constant (k_{out}), which yielded baseline values of 2.14 and 3.27 mg L^{-1} for TNF-α and IL-8, respectively. For IL-6, due to the high within-patient variability shown in the placebo group, the associated baselines were estimated independently from k_{in} and k_{out} with an estimate of 0.695 mg L^{-1}. A time-dependent k_{in} was introduced to capture the varying IL-6 secretion rate over time. The initial IL-6 secretion rate was estimated to be 1.86 mg L^{-1} h^{-1} with an exponential decay factor ($k_{decline}$) of 0.038 h^{-1}, which indicates that the secretion of IL-6 is stimulated to a larger degree at the beginning of the LPS challenge compared to later time points during the challenge.

To account for the observed time delay between LPS exposure and the increased secretion of cytokines, DDEs were applied and the delay factors (τ) were estimated to be 0.924, 1.46 and 1.48 h for TNF-α, IL-6 and IL-8, respectively. The extents of the increased cytokine secretion were shown to be related to LPS in a dose-dependent manner. Linear relationships of the stimulus of LPS on k_{in} were found to best capture the data compared to the other tested functions. The slope factors quantifying the stimulatory effect of LPS (S_{LPS}) were estimated to be 52.8, 41.5 and 67.1 L mg^{-1} h^{-1} for TNF-α, IL-6 and IL-8, respectively.

3.3 | C-reactive protein dynamics

The secretion of CRP was assumed to be exclusively stimulated by IL-6.[17] The developed IL-6 model with fixed parameters was used to generate IL-6 profiles in the fit of the CRP sub-model.

The baseline, secretion and degradation rate of CRP were estimated separately. The final estimates of baseline, k_{in} and k_{out} for the CRP model were 0.381 mg L^{-1}, 0.00095 mg L^{-1} h^{-1} and 0.0183 h^{-1}, respectively, along with an time delay factor of 4.2 h, and a slope factor of 19.2 L mg^{-1}. Separate residual error models were tested to distinguish the error associated with the different assays applied for CRP quantification in Studies B and C. Due to the similarity in the

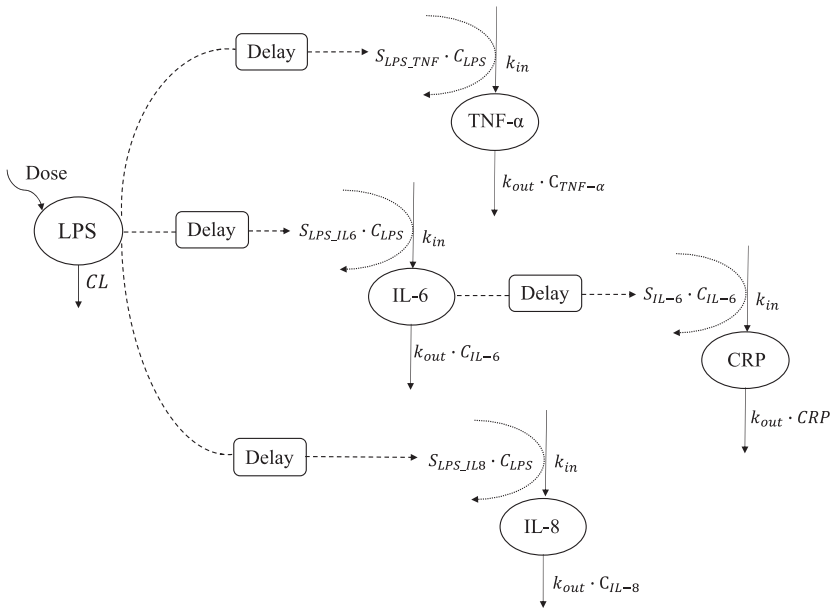

FIGURE 1 Developed model structure for inflammatory marker dynamics after lipopolysaccharides (LPS) challenge. LPS in plasma stimulates the release of cytokines tumour necrosis factor α (TNF-α), interleukin 6 (IL-6) and interleukin 8 (IL-8), while IL-6 induces the production of C-reactive protein (CRP). Delays were captured using delay differential equations with an explicit delay parameter τ

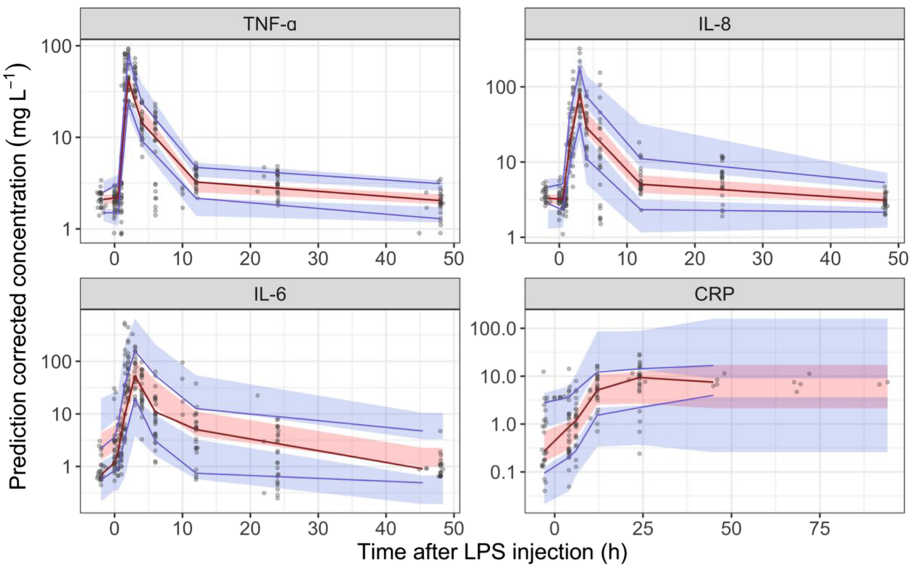

FIGURE 2 Stratified prediction corrected visual predictive checks based on 1000 simulations for tumour necrosis factor α (TNF-α), interleukin 8 (IL-8), interleukin 6 (IL-6) and C-reactive protein (CRP) after lipopolysaccharides (LPS) challenge. Solid lines are the 10th (lower), 50th (middle) and 90th (upper) percentiles of observations (indicated by symbols). The shaded areas are the corresponding 95% (2.5–97.5%) confidence intervals of the prediction percentiles

estimated proportional error for the residual unexplained variability in these two studies (31% for Study B *vs* 33% for Study C), a parsimonious model was selected, using the same proportional error for both studies (Table 2).

4 | DISCUSSION

We developed a novel modelling framework which successfully characterized the dynamics of multiple inflammatory markers following an LPS challenge in healthy volunteers. To our knowledge, this study is the first to quantitatively characterize inflammatory marker dynamics in humans after an LPS challenge.

Previous efforts developed models to quantify the inflammatory marker dynamics after LPS administration in a number of animal species.[10,11] In these preclinical studies, IDR models were used to describe cytokine dynamics in piglets and rats, which showed that system-specific parameters including secretion rate (k_{in}) and degradation rate (k_{out}) vary among species. For example, the k_{out} for TNF-α was estimated to be 2.04 h^{-1} in rats,[11] while it was approximately half (0.96 h^{-1}) in piglets.[10] In the current human in vivo model, we estimated a k_{out} of 0.36 h^{-1} for TNF-α, which is lower than in the previously mentioned piglet study. The observed trend of decreasing k_{out} with increasing body size was not observed for IL-6 (Figure S5), where the estimated values were 0.436, 3.45 and 0.305 h^{-1} for rat, piglet and human, respectively. Apart from our study, modelling work for IL-8 is still limited in both animal and human studies with LPS challenge. As no effects of LPS were included on k_{out}, this parameter could hold potential for aiding translation between species. Applying our developed modelling framework to both animal data and patient

TABLE 2 Final parameter estimates of the dynamic models for inflammatory markers upon lipopolysaccharide stimulation

Parameters (unit)	Parameter description	Parameter values (RSE)			
		TNF-α	IL-8	IL-6	CRP
Population parameters					
BL (mg L^{-1})	Baseline value	2.14 (3.7%)	3.27 (4.7%)	0.695 (19%)	0.381 (27%)
S (L mg^{-1} h^{-1})	Stimulation effect factor	52.8 (9.2%)	67.1 (20.3%)	41.5 (17.9%)	19.2 (51.7%)
k_{in} (mg L^{-1} h^{-1})	Production rate	-	-	1.86[a] (28.5%)	0.000950 (47.6%)
k_{out} (h^{-1})	Degradation rate	0.357 (5.7%)	0.320 (13.7%)	0.305 (23.5%)	0.0183 (20.3%)
$k_{decline}$ (h^{-1})	Decay factor for production rate	-	-	0.0380 (11.8%)	-
τ (h)	Time delay	0.924 (0.8%)	1.48 (0.2%)	1.46 (0.3%)	4.20 (1.4%)
Inter-individual variability					
BL		0.0270 (40.1%)	0.0350 (45.6%)	0.587 (65.6%)	1.70 (35.5%)
S		0.140 (40.9%)	0.655 (35.3%)	0.483 (122.4%)	1.09 (38.4%)
$k_{in}0$[a]		-	-	1.98 (23.7%)	-
k_{out}		0.0350 (65.8%)	0.262 (42.4%)	1.32 (27%)	-
Covariance η_S – η_{Kout}		-	−0.341 (14.1%)	-	-
Covariance η_{kin0} – η_{Kout}		-	-	1.37 (38.7%)	-
Residual variability					
Proportional		6.20% (9.3%)	12.3% (10%)	57.6% (5.8%)	31.6% (8.4%)

Abbreviations: RSE: relative standard error; TNF-α: tumour necrosis factor α; IL-8: interleukin 8; IL-6 interleukin 6; CRP: C-reactive protein.
[a]For IL-6, $k_{in}0$ instead of k_{in} was estimated as the initial production rate according to Equation (3): $k_{in} = k_{in}0 \cdot e^{(-k_{decline} \cdot t.)}$

data allows the quantification of between-species and/or -population differences, which could ultimately address the translational gap between preclinical animal models, healthy human volunteers and patients.

We modelled a stimulatory effect of IL-6 on CRP production. This could be supported by previous studies that described a central stimulatory effect of IL-6 on the release of hepatic acute phase proteins such as CRP in both animal[20] and human studies.[21] CRP is a useful marker used to monitor response to treatment,[5] but it has a long half-life of 19 hours. Upon an inflammatory stimulus, CRP usually starts to rise after 4–6 hours, reaching a maximum concentration after 36–50 hours.[5] This results in relatively slow dynamics of CRP in systemic inflammation, including sepsis, and makes it an issue in the current intensive care management of patients. The identified relationship between IL-6 and CRP with responsive kinetic characteristics suggests that IL-6 may be a more suitable biomarker for outcome prediction and to inform treatment optimization. This is supported by a recent study, in which IL-6 was found to be better than procalcitonin (PCT) and CRP in predicting the treatment success in predominantly non-surgical sepsis in the first 48–72 hours.[22]

In our analysis, the final estimate of k_{out} for CRP in healthy volunteers was 0.0183 h^{-1}, which is slower than an estimated value of 0.05 h^{-1} in neonatal sepsis patients under treatment with teicoplanin.[23] This might be related to the immature liver and renal function of neonates, since the liver is the main producer of CRP and renal function could influence its elimination. Meanwhile, our estimate is similar to an estimated value of 0.0238 h^{-1} based on pooled data from both healthy volunteer studies and patient studies with autoimmune diseases,[24] which could support the previous suggestions that CRP elimination rate is relative constant in adults under all conditions of health and disease.[5] However, the k_{in} of CRP in our study was estimated with relatively large RSE% (47.6%), which can be explained by the scarcity in data informing the elimination phase, where only four subjects were sampled up to 96 hours with only two samples after peak time post LPS administration. Meanwhile, the IL-6 and CRP measurements of these four subjects in Study C were shown to be higher compared with subjects in Study B (Figure S2), which can explain the observed underprediction of the population predictions due to the larger sample size of Study B. The lack of CRP peak concentration data, the different measurement assays and available sampling size for Studies B and C, and the lack of LOQ information, might have impacted the accuracy of the CRP-associated parameter estimates.

In comparison to previous preclinical models,[10,11] which applied E_{max} models to characterize the nonlinear LPS exposure–cytokine response relationships, we could only identify linear relationships. This is likely due to the modest LPS exposure levels attained in this study. To date, various LPS doses (0.06–4 ng kg^{-1}) have been used to replicate different inflammation conditions in healthy volunteers.[25] With the limited dose range applied in our study (0.5–2 ng kg^{-1}), exploring wider dose ranges and prolonged exposures of LPS in future studies could potentially help identify a nonlinearity and tolerance phenomena in the LPS exposure–response relationships.[10,26] In addition, complementary preclinical studies were more likely to characterize such relationships due to the higher flexibility in dose ranging in comparison to healthy volunteer studies.

Correlations between cytokines were also evaluated in previous studies.[2,6,10,27] TNF-α was shown to induce the IL-8 expression in human micro vessel endothelial cell lines,[28] while a stimulatory effect of TNF-α on IL-6 production was established in piglets.[10] In our study, a simultaneously estimated model including all the inflammatory biomarkers was tested to evaluate correlations between cytokines. However, due to the limited dosage range and sample size in the pooled studies, correlations could not be accurately estimated. As a result of estimation issues related to these correlations, and the overall decreased model stability, the simultaneous model was deemed inferior to the separately estimated models with no correlation between cytokines.

Time delays between LPS exposure and release of cytokines have been observed in both animal and human studies.[10,13] These delays are assumed to be a result of signal transduction of the LPS/TLR4 pathway[29] for cytokine synthesis and release.[30] Transit compartment models (TCMs) have been applied to capture such delay phenomena in the previous preclinical LPS challenge studies.[10] In our study, we found that DDEs outperformed TCM in capturing the time delay and baselines especially for CRP. When fitting TCMs, there was consistent overestimation of baseline values, regardless of the number of transit compartments and whether or not we estimated degradation rate (k_{out}) separately from transit rate (k_{tr}). Additionally, in comparison to TCM, DDEs can be numerically easier to fit due to the use of a single numeric delay parameter and they are more clearly interpretable than the mean transit time between transit compartments.[31]

Three cytokines and one acute phase protein were included in our quantitative framework based on human healthy volunteer studies. However, there are also other inflammatory markers which may play a crucial role in TLR4-mediated inflammation, like other pro/anti-inflammatory cytokines,[2] complement[32] and coagulation factors,[33] matrix metalloproteinases,[34] adhesion molecules[35] and other acute phase proteins.[36] These markers could be included in future studies to gain a more holistic and quantitative characterization of relevant inflammatory response.

In conclusion, we successfully developed quantitative models to capture the relationship between LPS and the cytokines TNF-α, IL-6 and IL-8, as well as between IL-6 and CRP in healthy human volunteers. This work could constitute the first step towards a more comprehensive model-based framework of immune-system response after LPS challenge in healthy volunteers. Such a framework could aid in further quantitative interpretation of inflammation marker dynamics among species, which is the basis for inter-species translation of findings in drug development in the field of systemic inflammation.

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COMPETING INTERESTS

The authors declare no conflicts of interest.

CONTRIBUTORS

F.L., M.M. and J.G.C.H. designed the study; F.L. and T.G. performed the data analysis; L.B.S.A., T.G., E.H.J.K, M.M., P.H.G. and J.G.C.H. supported interpretation of the results; F.L., L.B.S.A. and J.G.C.H. wrote the paper; J.G.C.H. conceived the project; all authors reviewed the paper.

DATA AVAILABILITY STATEMENT

The modelling codes supporting the findings of this study are available within the supplementary information files.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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