

A Multiscale Computational Model Predicts Human Liver Function From Single-Cell Metabolism

König M.¹, Marchesini G.², Vilstrup H.³, Somogyi A.⁴ and Holzhütter HG.¹

¹Department of Computational Systems Biochemistry, University Medicine Charité Berlin, D-10117 Berlin, Germany

²Department of Internal Medicine, Aging and Nephrological Diseases, University of Bologna, I-40138 Bologna, Italy

³Department of Hepatology and Gastroenterology, Aarhus University Hospital, DK-8000 Aarhus, Denmark

⁴Biocomplexity Institute, Indiana University, Simon Hall MSB1, Bloomington, IN 47405

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To whom correspondence should be addressed: Matthias König, Institute of Biochemistry, University Medicine Charité Berlin, Virchowweg 6, 10117 Berlin; Tel: (0049) 30450528197; Email: matthias.koenig@charite.de

ABSTRACT

Understanding how liver function arises from the complex interaction of morphology, perfusion, and metabolism requires the application of computational approaches from the cellular level up to the entire organ.

We report a multiscale mathematical model of the Human liver bridging the scales from cellular processes over tissue representation of ultra-structure and micro-circulation to the entire organ integrated with perfusion. The model integrates heterogeneous data on multiple spatial and temporal scales and was validated against a broad range of data across multiple scales.

Herein we describe the model application to hepatic galactose metabolism and to liver function tests based on galactose elimination capacity (GEC). Major results are i) the personalization of the multiscale model resulting in improved evaluation of liver function tests (GEC), ii) explanation of changes in liver function with aging due to perfusion and volume changes of the liver, and iii) prediction of population variability in liver function based on variability in liver volume and perfusion.

We conclude that physiology- and morphology-based multiscale models can improve the prediction of individual liver function.

INTRODUCTION

About five million individuals are affected by liver diseases in Germany. The causes of these diseases often remain unknown, despite the enormous progress made in terms of their treatment. The elucidation of the fundamental mechanisms underlying liver diseases and the development of new treatment approaches are therefore of great importance and, at the same time, they pose a great challenge to the public health care system.

The liver is the metabolic center of the body performing hundreds of functions, including storing nutrients; maintaining the homeostasis of numerous plasma metabolites; detoxification of xenobiotics; and the clearance of substances such as galactose.

Prevalence liver disease ?

The causes of liver diseases despite enormous progress in their treatment often remain unknown.

Therefore, the elucidation of underlying principles how liver function results of a complex interaction between morphology, perfusion and metabolism is of great importance.

Biological systems like the liver are inherently complex in nature; they are composed of multiple functional networks that operate across diverse temporal and spatial domains {Walpole2013}.

Computational models are uniquely positioned to capture the connectivity between these divergent scales, as they can bridge the gap in understanding between isolated in vitro experiments and whole-organ in vivo models {Walpole2013}.

Liver architecture

The liver has a unique architecture consisting of a multitude of structural similar hexagonal subunits connected in parallel, the so-called lobuli. Within a single lobule a network of capillaries, the liver sinusoids, connect the outer periportal regions, supplied with blood via portal vein and hepatic artery, with the central perivenous region, drained by the central vein {Sasse1992}. The sinusoids, separated from the surrounding hepatocytes via the space of Disse, form the smallest functional unit of the liver (Figure 1).

GEC as Liver function test

Quantification of liver function is necessary to assess the degree of liver impairment, to evaluate response to treatment and to select transplant recipients {Dufour1992}. The liver is the primary organ for clearance and whole-body metabolism of galactose {Bernstein1960, Berry2000, Segal1971} making the rate with which a given plasma load of galactose is cleared a liver-specific function test. Hepatic galactose elimination follows Michaelis-Menten saturation kinetics {Keiding1973, Keiding1976} with a concentration-dependent (first-order) elimination phase at low galactose concentrations and a definable clearance maximum, the galactose elimination capacity (GEC), at higher concentrations (zero-order phase) {Schirmer1986}. Important determinants in galactose clearance are hepatocyte function and liver blood flow, with low galactose clearance measuring the estimated hepatic blood flow, whereas high galactose concentration measures the hepatic functional capacity. Determination of the galactose elimination capacity (GEC) {Marchesini1988, Schirmer1986, Tygstrup1966} is established tests

of liver function measured in numerous studies {[Jepsen2009](#), [Fabbri1996](#)}. Impairment of the liver by diseases like cirrhosis {[Henderson1982](#), [Jepsen2009](#)} or intoxication {[Vilstrup1983](#)} or reduction in liver volume/parenchyma like in partial hepatectomy {?} or CCL4 intoxicification {?} is commonly associated with reduced galactose clearance.

Hepatic Galactose Metabolism

Galactose is metabolized by hepatocytes via three key enzymatic steps: i) the phosphorylation of galactose (gal) to galactose 1-phosphate (gal1p) catalysed by galactokinase (GALK, EC 2.7.1.6); ii) the conversion of gal1p to UDP-galactose (udpgal) by galactose-1-phosphate uridyl transferase (GALT, EC 2.7.7.10) and iii) the interconversion of udpgal and UDP-glucose (udpglc) by UDP-galactose 4'-epimerase (GALE, EC 5.1.3.2) {[Novelli2000](#), [Petry1998](#)}. Galactose can enter glycolysis as glucose-1 phosphate (glc1p), one of the GALT reaction products, or can be incorporated as udpgal, the substrate donor of all galactosylation reactions, in glycoproteins and glycolipids {[Novelli2000](#)}.

Despite the importance of the hepatic galactose metabolism for the systemic galactose clearance and formation of precursors for glycosylation reactions no detailed mathematical model of this pathway is available hitherto.

Liver Function in Aging

The percentage of deaths attributed to liver disease increases dramatically in humans beyond the age of 45 years {[Schmucker2005](#)}. Aging is characterized by normal progressive decline in functions that, cumulatively, diminish a cell's, organ's or organism's capacity to respond to intrinsic or extrinsic stimuli. Physiologic changes known to play a role in the pharmacokinetics of a drug in the elderly include: decline in total body mass, liver volume, liver blood flow and liver function {[Anantharaju2002](#), [Wynne1989](#)}. In addition, the ultrastructure of the liver changes with aging: Pseudocapillarization, characterized by defenestration, thickening of the endothelium, and deposition of basal lamina and extracellular matrix in the space Disse, is a widespread change in aging {[McLean2003](#), [LeCouteur2001](#), [Cooger2003](#)}. The fenestrated endothelial cells of the liver sinusoids act as a dynamic filter that permits exchange of fluid, solutes and particles between the sinusoidal lumen and space of Disse {[Cogger2003](#)}. Age-related changes in the liver sinusoids and space of Disse are implicated in the association between ageing and impaired clearance of drugs {[LeCouteur1998](#)} and may provide a mechanistic link between primary aging processes and age-related disease {[LeCouteur2002](#), [Cogger2003](#)}.

Multiscale-Model

Multiscale computational approaches are required to elucidate the complex interaction of organ structure, perfusion, and metabolism. Insights in how the successive dilution or concentration of metabolites along the sinusoid from the periportal to the perivenous site influences cellular metabolism requires a tissue-scale model that combines spatial gradients of metabolite concentrations with the cellular metabolism of individual liver cells. Finally, for the correct interpretation of results obtained in the galactose tolerance test it is necessary to take into account regional variations of the blood flow and in the architecture of sinusoids. Thus, for fully

understanding the metabolic input-output relationship of the liver it needs to bridge the spatial scales from single hepatocyte metabolism over tissue-scale functional units of sinusoids to the whole-organ level.

Furthermore, for the individualized evaluation of liver function it is necessary to personalize the model based on anthropomorphic information like age, gender, bodyweight and height.

Systems-level approaches are required to analyze the combined effects of alterations in perfusion, liver volume and ultrastructure like occurring in aging and their effects on liver function tests like GEC. This has important consequences for the evaluation of the functional capacity/reserve and the detection of impairments and disease, both crucial for organ evaluation in transplantation or in the calculation of proper drug dosage depending on age.

The objective is to develop a multiscale computational model which describes physiology, morphology and function of the human liver.

Here we present a multiscale model of the human liver ...

Our model attempts to (1) describe ... ; (2) account for ...; (3) accurately predict a wide range of observable liver behavior;

RESULTS

We present a multiscale mathematical model bridging the scales from individual cellular processes to the level of the entire organ. The model can be used to describe physiology, morphology and function of the human liver by integrating hepatic metabolism with perfusion and liver structure (Figure 1). The model combines detailed kinetic models of cellular metabolism with a tissue-scale perfusion model of the sinusoid. The cellular model is integrated into a realistic tissue-scale model of the sinusoidal liver unit based on known histological parameters (geometry, cell numbers, architecture, perfusion rates). Regional liver metabolism was modelled as weighted average across the heterogeneous contributions of tissue-scale models with differing in blood flow rates and tissue structure. Finally, the function of the entire organ is modelled by scaling of the regional output based on individual liver perfusion and liver volume, resulting in individualized liver function.

The model integrates and predicts experimental data on multiple temporal and spatial scales: i) multiple indicator dilution curves; ii) extraction and clearance of galactose depending on perfusion and galactose load; iii) individual GEC, population variability in GEC and the dependency of GEC on aging; iv) classification of liver disease based on predicted GEC. Personalized multiscale models are generated based on individual anthropomorphic information in combination with fitted relationships describing the dependencies of hepatic volume and blood flow on these features. We demonstrate possible clinical application of the presented systems biology approach by outperforming regression approaches in the classification of liver disease in a retrospective analysis of a large cohort study.

Model Construction

Our approach to developing was ...

Mean sinusoidal unit & the integrated response over the heterogeneous contributions of sinusoids based on heterogeneity in ultra-structure and microcirculation are presented.

Multiple Dilution-Indicator Curves

In a first step the model was validated multiple-indicator dilution curves (Figure ?) {Goresky1973}. The single-injection, multiple-indicator dilution approach provides a method to determine the composition of the liver and the rates of hepatic processes {Goresky1973}. Labeled red blood cells (RBC) are used as vascular reference. Larger materials are excluded from the space of Disse. The outflow concentration of each tracer is divided by the total injected, providing a normalized value, an outflow fraction per ml.

At low blood galactose concentrations, the labeled galactose appears at the outflow with labeled sucrose, but is much reduced in magnitude, and exhibits a long tailing. Its outflow recovery is much reduced. At high blood galactose concentrations, the initial part of the profile increases towards that for labeled sucrose, the tailing becomes much larger in magnitude, and the outflow recovery becomes virtually complete {Goresky1973}. In addition the altered galactose tracer peaks under varying galactose background provides testing of the model.

The shift of the dilution-indicator curves observed and explained by Goresky as different free volumes of the substances are a consequence of the different diffusion coefficients for the substances, resulting in a delayed appearance of the substances perivenous.

None of the data was used for model fitting, all model parameters result from the geometric constraints of the system and the physio-chemical properties of the substances transported within the sinusoid and the space of Disse.

Lobulus Heterogeneity

Extreme heterogeneity on the lobulus & sinusoidal units (network for clearance, broad spectrum of parameters)

The integrated behavior based on molecular detailed description of single cell behavior give the correct multiple dilution indicator curves, consequently describing correctly the distribution of substances in the various spaces.

Multiscale-model provides predictions about the cell to cell variability and the heterogeneity within the lobulus.

Not only important gradient along sinusoid, but strong variation from sinusoid to sinusoid depending on the flow conditions.

Galactose Elimination Capacity

Via integration of the heterogenous sinusoidal units the hepatic galactose clearance, extraction ratio (ER) and the galactose elimination capacity for given perfusion per volume liver are calculated. The model reproduces the observed saturation in galactose clearance, the dependency of the extraction ration of galactose on the perfusion.

Hepatic galactose elimination follows Michaelis-Menten saturation kinetics {Keiding1973, Keiding1976} with a concentration-dependent (first-order) elimination phase at low galactose concentrations and a definable clearance maximum (GEC) at higher concentrations (zero-order

phase) {Schirmer1986}.

Galactose Clearance at low concentrations has proven to be as near ideal a method for estimating the effective hepatic blood flow (EBHF) because most of the plasma galactose entering the liver also leaves the liver without being metabolized {Schirmer1986}. Henderson et al. confirmed the near complete extraction of galactose across the liver (ER = 0.94) in subjects without hepatic disease by performing hepatic vein catheterization {Henderson1982}.

The most extensive work on galactose elimination kinetics was done by Keiding and co-workers {} [43-45, 48, 50, 85, 99]. "These kinetic studies on the clearance of galactose at concentrations of 0 to 10 mg/dl (0 to 0.555 mmol/l) show that it approaches the ideal test substance for measuring effective liver blood flow (EHBF) (a) it is kinetically simple to analyse at steady state during continuous infusion (b) it is avidly removed by the liver, with minimal extrahepatic clearance and (c) there is indirect evidence supporting virtually complete extraction by functional liver tissue on each pass.

High galactose (GEC) Under high galactose concentrations like occurring in the galactose clearance tests the model predicts a capacity limited galactose clearance, thereby measuring the galactose elimination capacity. Hepatic vein catheterization at high concentrations revealed a constant hepatic arterial-hepatic venous concentration difference {Tygstrup1954, Tygstrup1958}. Since all hepatocytes are thought to participate maximally in the removal of galactose, the test has been regarded as a measure of the functioning liver cell mass (Lm) {Tygstrup1966}.

Elimination Rate, Elimination Fraction

Flow dependency

15 - 9 - 15 ml/min (perfusion ?) "There was significantly lower outflow concentration during the period with low flow than during periods with high flow. {Keiding1978} -> flow dependency at low concentrations"

Individual GEC prediction

GEC in aging A significant negative correlation as observed between age and both liver volume and apparent liver blood flow above 30 years. The reduction in liver volume, apparent liver blood flow and perfusion may at least partly account for the decline in the clearance of many drugs undergoing liver metabolism, which has been noted to occur with aging in man {Wynne1989, Schnegg1986}. Also during childhood major absolute and relative changes per body weight occur in liver volume and bloodflow. To test if this underlying changes in liver morphology and perfusion can explain the age-dependent changes

Application of the model revealed that variability of galactose clearance in aging is mainly explained by changes in liver structure, perfusion and morphology during lifetime. These alterations with age have important implications for drug dosing.

(GEC ~ liver volume in rats during aging , Desktop Nielsen-Fischer, Keiding)

Population Variability Given a cohort with anthropomorphic features for the individuals our model allows the prediction of expected distribution of GEC values for the individuals in the cohort. If the cohort is representative for the population, the population variability of liver function can be estimated. We estimate the population variability in the US population based on the NHANES cohort {NHANES} (Figure 5). Not only GEC and GECkg are predicted correctly, but other pairwise correlations like the dependency of liver volume and blood flow from age, bodyweight, height and BSA (see Figures supplement). The presented methods allows thereby an estimation of the distribution of liver function based on the variability of hepatic perfusion and liver volume in the population.

GEC classification

The predicted distribution of liver function (GEC) for an individual can be employed for an improved evaluation of the result of a liver function test. I.e. if the further the measured GEC from the expected GEC, the higher the probability for liver disease. This approach was evaluated retrospectively in a large cohort study {Fabbri1996} and with data available from the literature (see table ?), which measured GEC in combination with anthropomorphic information. A classifier based on our predictive GEC model outperformed classical logistic regression between GEC and liver disease, even when age and bodyweight were included in the logistic regression.

The GEC prediction in combination with the classifier for liver disease was implemented in a web application allowing the presented calculation of GEC range and classification of liver disease based on the presented model.

Improved liver function tests

We demonstrate the application of a personalized multiscale model of the human liver providing improved evaluation of an established liver function test.

DISCUSSION

We have developed a multiscale. model that accounts for ... and explains a variety of emergent behaviors in terms of molecular interactions.

Our model accurately recapitulates a broad set of experimental data

provides insights into several biological processes for which experimental assessment is not readily feasible, and enables ...

The model includes the common key processes that lead to liver diseases, metabolism, perfusion and ultrastructure of the liver.

Metabolic changes & gene expression, individual levels of metabolism

Galactose metabolism and GEC are quite constant. Adult rats fed a 40% galactose diet for 5 days did not show an increase in GEC although 20 days on the diet resulted in a 20% increase in V_{\max}

suggesting that adaptive mechanisms are slow [Schirmer1986 ->18]. This lack of inducability and relatively constant V_{\max} is desirable in clearance methodology as a fluctuating V_{\max}/FK_m would certainly complicate clearance interpretations.

Model assumptions

A mathematical model is always only a selective representation of reality. The results have to be evaluated in the context of the underlying assumptions of the model. Many of these assumptions are due to a lack of experimental data.

- For instance, the distributions of sinusoidal parameters were assumed independent from each other. As soon as correlation data becomes available the model can be updated.
- Dispersion of dilution peaks in the large vessels and runtime differences were not modelled. Relationship vascular tree and sinusoid transit times ? It is assumed that no displacement occurs between reference intravascular and diffusible tracers in the large vessels: all displacement occurs in the exchanging vessels (sinusoids). The interrelations between whole-organ outflow reference and diffusible tracer curves will depend not only on the phenomena occurring within each sinusoid but also on the way the transit times in larger vessels and sinusoids are interrelated. Various combinations are possible, depending on the structure of the network and the kind of flow coupling in the system. The pattern corresponding to the liver was found to lie at a simple extreme in this possible spectrum [Rose1976, Goresky1970]. The distribution of out-flow transit times was found to correspond to the distribution transit of sinusoidal times in large transit times; the distribution of vessels was so compact that a single value could be assumed. Thus it was possible to derive a test for the single-sinusoid modeling. If, after a common transit time in large vessels, the sinusoidal transit time for each diffusible label in the liver is increased by the ratio of its total-to-accessible sinusoidal vascular space, then it should be possible to reverse this flow-limited delay effect in the curve for each diffusible label.
- Goresky et al.1 previously have considered two models representing the extreme cases, i.e., no heterogeneity, and maximum heterogeneity in capillary transit times. Multiple indicator-dilution data from the liver fit the latter model well [Rose1976].
- Heterogeneity in local blood flow in the liver was not taken into account.

Heterogeneity

Could explain the observed heterogeneity observed in NAFDL (locally different concentrations).

Comparison to current multiscale models of liver

Höhme (no detailed blood flow, no metabolism)

Chaloubh (missing parameter distributions, only bridging the gap to the sinusoidal unit, not

possible to simulate the different effects of heterogeneous variations of parameters.

Galactose-Clearance Modelle – no detailed description of metabolism, no bridging to whole liver function

Ricken, porous media

Selection bias

The prediction of individual liver volumes & flows is based on selected available studies for the correlation. The predictions reflect this subset of data used for model fitting. Care was taken only to use data for Caucasian/Western individuals with normal bodyweight range and without any liver disease.

Improved function tests

‘Reference ranges play an important role in clinical medicine, with values that lie outside the reference range viewed as an indication for further investigation and/or treatment.’ {Cole2009}

The mechanistic parameters, namely liver volume and hepatic blood flow were described with probability density functions (PDF) estimated from individual subject data (LMS, GAMLSS) approach. Since each PDF depicts the frequency of occurrence of all expected values for each parameter in the population, the effects of multiple sources of uncertainty and variability were accounted for in the estimated distribution of GEC in the population.

Towards a virtual liver & future applications

Bridging the scales from cellular processes over the coupling of single cells within the tissue-architecture towards whole-organ models is a crucial step in understand physiological function of organs in the normal state and in pathophysiology. Only by modelling the different scales explicitly the emerging behaviour on a liver scale can be properly understood.

the systems biology approach, i.e. the interaction between biological experiments and mathematical modelling, is to be transferred to application-oriented liver research as a next step

In order to use the understanding of these processes to develop novel treatment and prevention approaches, disease-relevant and, if possible, personalized multiscale models are to be derived.

Earlier detection of liver disease, earlier intervention

ONLINE METHODS

The presented liver model is a multi-scale model comprising cellular scale on the level of single hepatocytes (Figure 1A), tissue scale on level of the sinusoidal unit (Figure 1B), a region of interest of the liver (lobulus, Figure 1C) and the liver in individual subjects and variability in the population (Figure 1D).

Availability of data and models

The complete source code of modelling and analysis is open source and available under GPL from <https://github.com/matthiaskoenig/multiscale-galactose>. All literature data sets are made accessible on request. The mathematical model on cellular and tissue-scale are published as SBML under creative commons (CC BY-SA 4.0) in the Supplementary Information as well as on Biomodels.org and JWS Online. A HTML representation of the information is available in the Supplement.

Numerical integration

The hepatocyte and sinusoidal unit models are kinetic model based on ordinary differential equation (ODE) systems. All numerical integrations were performed with libRoadRunner {Somogyi2014} with absolute and relative tolerances of 1E-6 on a computer cluster. libRoadRunner was extended to efficiently handle the large models via ...

All results were stored in a database.

Cellular scale - galactose metabolism

The kinetic model of galactose metabolism for individual hepatocytes consists of three main enzymatic steps i) the phosphorylation of galactose (gal) to galactose 1-phosphate (gal1p) catalysed by galactokinase (GALK, EC 2.7.1.6); ii) the conversion of gal1p to UDP-galactose (udpgal) by galactose-1-phosphate uridyl transferase (GALT, EC 2.7.7.10) and iii) the interconversion of udpgal and UDP-glucose (udpglc) by UDP-galactose 4'-epimerase (GALE, EC 5.1.3.2) {Novelli2000, Petry1998}. Galactose can enter glycolysis as glucose-1 phosphate (glc1p), one of the GALT reaction products, or can be incorporated as udpgal, the substrate donor of all galactosylation reactions, in glycoproteins and glycolipids {Novelli2000}. The alternative processes important in galactosemias and ATP synthesis (ATPS) and NADP reduction (NADPR) for cofactor regeneration were added to the model. Detailed information on metabolites, initial concentrations, rate equations and enzymatic parameters is provided in Supplementary Table 1 and Supplementary Table 2. All literature based kinetic parameters were included in SABIO-RK {Wittig2012} (see Supplementary Tables and SBML annotations). Maximal enzyme activities (V_{\max}) were chosen to achieve good correspondence of model simulations with reported galactose elimination rates in young subjects (20 years).

Tissue scale - sinusoidal unit

The tissue-scale model of the sinusoidal unit (Figure 1B) consists of a central blood vessel (sinusoid) surrounded by the space of Disse and adjacent hepatocytes in cylindrical geometry

with parameters in [Supplementary Table 3](#) and [Supplementary Table 4](#). The periportal (pp) and perivenous (pv) blood compartments are located adjacent to the first and last sinusoidal volume, respectively. A single sinusoidal unit consists of N_c hepatocytes with each cell having a single associated sinusoid and Disse volume. In the sinusoid substances are transported by blood flow and diffusion, in the space of Disse solely by diffusion. Red blood cells (RBC) are constricted to the sinusoid, whereas all other model substances smaller than the fenestrae ($r_{\text{substance}} \leq r_{\text{fen}}$) pass in the space of Disse owing to the fenestration of the endothelial cells [{Wisse1985}](#), i.e. galactose, albumin, sucrose and water. Galactose and water are exchanged between the space of Disse and the hepatocytes, whereas sucrose and albumin are restricted to the space of Disse.

Diffusion and blood flow are modelled via discretized one-dimensional diffusion and convection equations (analogue to [{Konig2013}](#)). The diffusion through the sinusoidal fenestration, small cylindrical channels in the endothelial cells is described via pore theory [{Pappenheimer1953, Renkin1954}](#). The total restriction to diffusion due to the combined effects of steric hindrance at the entrance of the pores and frictional resistance within the pores for substance a with radius r_a is given as actual diffusion D_a relative to unhindered Diffusion $D_{a,0}$ with radius of the substance r_a and pore radius r_{fen} as $\frac{D_a}{D_{a,0}} = (1 - \frac{r_a}{r_{\text{fen}}})^2 \left[1 - 2.104 (\frac{r_a}{r_{\text{fen}}}) + 2.09 (\frac{r_a}{r_{\text{fen}}})^3 - 0.95 (\frac{r_a}{r_{\text{fen}}})^5 \right]$ [{Renkin1954}](#).

Liver region of interest

The heterogeneity of sinusoidal units within a lobulus was modeled via Monte Carlo simulation of varying sinusoidal units based on experimental distributions for parameters of ultrastructure (sinusoidal length L_{sin} , sinusoidal radius y_{sin} , width space of Disse y_{dis} , hepatocyte sheet thickness y_{cell}) and microcirculation (sinusoidal blood flow v_{blood}). The output of a region of interest was calculated via integration of the results from $N=1000$ sinusoidal units. All parameter distributions were assumed log-normal and statistically independent of each other. Distributions of y_{sin} , v_{blood} and y_{cell} were fitted based on maximum-likelihood method for uni-variate distributions. For L_{sin} and y_{dis} the log-normal parameters were calculated from reported mean m and standard deviation std via $stdlog = \sqrt{\log(1 + \frac{std^2}{m^2})}$ and $mlog = \log(\frac{m^2}{m^2 + std^2})$. All parameters and references are given in [Supplementary Table 4](#).

For the variation of perfusion, i.e. blood flow per tissue volume, the distribution of blood flows was scaled via $p_{\text{scaled}}(v_{\text{blood}}) = p(f_{\text{flow}} * v_{\text{blood}})$ mit $f_{\text{flow}}=1$ corresponding to the experimental microcirculation.

Liver

To scale the output from region of interests to main things have to be taken into account. 1) The region of interests describe the parenchymal liver tissue. To account for whole liver function the non parenchymal volume of the liver, mainly consisting of large vessel volume, has to be taken into account. The parenchymal fraction of the liver is around 80% ($f_{\text{tissue}}=0.8$).

The relationship between tissue perfusion $P_{\text{tissue}} = \frac{Q_{\text{tissue}}}{V_{\text{tissue}}} = \frac{\sum_{i=1}^N Q_{\text{sinuit}}}{\sum_{i=1}^N V_{\text{sinunit}}}$ and liver perfusion $P_{\text{liver}} = \frac{Q_{\text{liver}}}{V_{\text{liver}}}$

is given via $P_{\text{liver}} = \frac{Q_{\text{tissue}}}{V_{\text{tissue}} + V_{\text{notissue}}} = (\frac{1}{2-f_{\text{tissue}}}) \frac{Q_{\text{tissue}}}{V_{\text{tissue}}} = (\frac{1}{2-f_{\text{tissue}}}) P_{\text{tissue}}$

Via the relationship for normal perfusion of 1.2ml/min/ml an necessary adaption of the microcirculation of $f_{\text{flow}}=0.3$ results corresponding to a mean sinusoidal flow velocity of 81µm/s.

This is still in the range of the experimentally obtained values. OPS values and microcirculation is taken on the surface of the liver, with partly larger arterial components and properly not representative of the whole liver.

Multiple indicator dilution curves

An indicator substances introduced into the blood flowing into the liver become dispersed in the effluent blood and the concentrations of the substances in the effluent blood form an indicator dilution curve {Goresky1973}. The rapid injection of labeled red blood cells (a vascular indicator), labeled sucrose and albumin (extracellular references), and labeled galactose under various galactose concentrations into the portal vein in combination with rapidly sampled venous blood were simulated {Goresky1973, Goresky1983} were simulated. For comparison with the experimental data the catheter and nonexchangeable vessel transit time t_0 was estimated from the time of first appearance of radioactivity above background levels in the experimental and simulated dilution curves. Integration of the single a region of interest for the periportal output was done via the volume flow weighted average of the individual sinusoidal units

$$c_{venous}^s(t) = \sum_{i=1}^N w_i c_{pp}^s(t) = \sum_{i=1}^N \frac{Q_{sinunit,i}}{\sum_{i=1}^N Q_{sinunit,i}} c_{pp}^s(t)$$

The model is a distributed model of flow based on parallel, non-interacting sinusoids joined at the venous terminus. The dispersion characteristics is due to the *a priori* incorporated experimental velocity and path length variations within the ensemble of sinusoids {Weiss1995}

Galactose Elimination

The galactose elimination (GE), the removal rate (R), the extraction ratio (ER) and the clearance (CL) were calculated from the blood flow Q_{tot} and the periportal and perivenous concentrations of galactose c_{pp}^{gal} and c_{pv}^{gal} using the following equations {Schirmer1986}

$$GE = (c_{pp}^{gal} - c_{pv}^{gal})$$

$$R = Q (c_{pp}^{gal} - c_{pv}^{gal})$$

$$ER = \frac{(c_{pp}^{gal} - c_{pv}^{gal})}{c_{pp}^{gal}}$$

$$CL = Q \frac{(c_{pp}^{gal} - c_{pv}^{gal})}{c_{pp}^{gal}}$$

For the region of interest the sinusoidal units are integrated resulting in

$$GE = (c_{pp}^{gal} - c_{pv}^{gal})$$

$$R = Q (c_{pp}^{gal} - c_{pv}^{gal})$$

$$ER = \frac{(c_{pp}^{gal} - c_{pv}^{gal})}{c_{pp}^{gal}}$$

$$CL = Q \frac{(c_{pp}^{gal} - c_{pv}^{gal})}{c_{pp}^{gal}}$$

Liver in aging

The age dependent change in total liver volume, blood flow and perfusion are taken into account in the age-dependent GAMLSS curves. The additional changes in ultrastructure of the liver

(pseudocapillarization) were modeled using the age-dependent change in fenestration, i.e. change in fenestration number per area (N_{fen}), and endothelial thickness (y_{end}) based on experimental data as input for the model predictions (supplementary information). Practically, for different combinations of (N_{fen} , y_{end}) corresponding to certain ages the GEC per tissue and perfusion were calculated.

Individualized predictions

Based on the integration of detailed kinetic models of sinusoidal units over a region of interest the metabolic function for given liver structure, morphology, perfusion and metabolic function can be calculated. The total hepatic function of a person results from this regional function with the actual liver volume and blood flow of the person. This is achieved via prediction of the individual liver characteristics based on the anthropomorphic data of the person, i.e. based on sex, age, bodyweight, height and body surface area.

In a first step generalized additive models for location, scale and shape (GAMLSS) {Stasinopoulos2007} were fitted to describe the correlations between single liver features and single anthropomorphic features based on individual data from >3000 subjects from >30 studies (supplement GAMLSS). Individual models for male, female and all data were fitted. The resulting models enable the prediction of the probability distributions of liver features for single anthropomorphic features based on gender, for instance the distribution of liver volumes depending on age for females $p_{sex=F, volLiver \sim age}$. Importantly, the observed population variability is part of the model.

In a second step, the information of the single feature models is combined under the assumption of statistical independence to generate a best estimate of liver volume based on the set of anthropomorphic features observed in the person. The result is a personalized probability distribution of liver volumes $p_k(volLiver)$ for the subject k with sex=S, age=A, bodyweight=B, height=H and bsa=BS

$$p_k(volLiver) = p(volLiver|sex = S, age = A, bodyweight = B, height = H, bsa = BS) =$$

$$\begin{aligned} & p_{sex=S, volLiver \sim age}(volLiver|age = A) \cdot \\ & p_{sex=S, volLiver \sim age, bodyweight=B}(volLiver|age = A, bodyweight = B) \cdot \\ & p_{sex=S, volLiver \sim age, bodyweight=B, height=H}(volLiver|age = A, bodyweight = B, height = H) \cdot \\ & p_{sex=S, volLiver \sim age, bodyweight=B, height=H, bsa=BS}(volLiver|age = A, bodyweight = B, height = H, bsa = BS) \cdot \\ & p_{sex=S, volLiver \sim age, bodyweight=B, height=H, bsa=BS, bodyweight=B}(volLiver|age = A, bodyweight = B, height = H, bsa = BS, bodyweight = B) \cdot \\ & p_{sex=S, volLiver \sim age, bodyweight=B, height=H, bsa=BS, bodyweight=B, height=H}(volLiver|age = A, bodyweight = B, height = H, bsa = BS, bodyweight = B, height = H) \cdot \\ & p_{sex=S, volLiver \sim age, bodyweight=B, height=H, bsa=BS, bodyweight=B, height=H, bsa=BS}(volLiver|age = A, bodyweight = B, height = H, bsa = BS, bodyweight = B, height = H, bsa = BS) \end{aligned}$$

Hepatic blood flows is calculated in a similar manner, but taking the additional correlation information between liver volume and blood flow into account. The resulting probability distribution of hepatic blood flow for person k with given liver volume V is

$$p_k(flowLiver|volLiver = V) = p(flowLiver|sex = S, age = A, bodyweight = B, bsa = BS) \cdot p_{sex=S, flowLiver \sim volLiver}(flowLiver|volLiver = V)$$

$$\begin{aligned} & p_{sex=S, flowLiver \sim age}(flowLiver|age = A) \cdot \\ & p_{sex=S, flowLiver \sim age, bodyweight=B}(flowLiver|age = A, bodyweight = B) \cdot \\ & p_{sex=S, flowLiver \sim age, bodyweight=B, height=H}(flowLiver|age = A, bodyweight = B, height = H) \cdot \\ & p_{sex=S, flowLiver \sim age, bodyweight=B, height=H, bsa=BS}(flowLiver|age = A, bodyweight = B, height = H, bsa = BS) \cdot \\ & p_{sex=S, flowLiver \sim age, bodyweight=B, height=H, bsa=BS, bodyweight=B}(flowLiver|age = A, bodyweight = B, height = H, bsa = BS, bodyweight = B) \cdot \\ & p_{sex=S, flowLiver \sim age, bodyweight=B, height=H, bsa=BS, bodyweight=B, height=H, bsa=BS}(flowLiver|age = A, bodyweight = B, height = H, bsa = BS, bodyweight = B, height = H, bsa = BS) \end{aligned}$$

Finally, the metabolic function of person k, in this case the galactose elimination capacity, is

calculated by scaling the metabolic function per tissue volume for given perfusion and structure, to total blood flow and liver volume ($flowLiver_k$ and $volLiver_k$) via the metabolic functions calculated for regions of the liver.

$$GEC_k = f_{GEC_per_volLiver}(flowLiver_k/volLiver_k) * volLiver_k$$

Via Monte Carlo simulation, i.e. repeated sampling from the individualized probability distributions $p_k(volLiver)$ and $p_k(flowLiver|volLiver)$ the distribution of liver volumes, blood flows and metabolic function in people with the given anthropomorphic features can be calculated.

Population variability

To calculate the population variability in liver function the prediction of liver volume, blood flow and GEC was performed for a large cohort representative of the US population. The NHANES {[NHANES](#)} survey data between years 1999 - 2012 was used, with subjects filtered based on body mass index ($18.5 \leq BMI \leq 24.9$) and ethnicity (Non-Hispanic White). For all subjects with complete data sets of age, gender, height, and body weight the prediction was performed. Using the Monte Carlo approach, repeated computations based on inputs selected at random from statistical distributions for each input parameter are conducted to provide a statistical distribution of the output. Using high percentile (e.g. 95th) and 50th percentile, the intraspecies variability can be calculated. To derive this information, Monte Carlo simulations based on distributions of input parameters have frequently been used. (Lipscomb et al., 2003; Gentry et al., 2002; Haber et al., 2002; Lipscomb and Kedderis, 2002; Timchalk et al., 2002; Bogaards et al., 2001; El-Masri et al., 1999; Thomas et al., 1996a, b).

Body surface area

Body surface (BSA) is calculated via DuBois formula from bodyweight and height $BSA = 0.007184 \cdot bw^{0.725} \cdot h^{0.425}$ {[Moesteller1987](#)}.

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