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Kinetic analysis of FDG in rat liver: Effect of dietary intervention on arterial and portal vein input

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Abstract

Introduction—Dietary conditions may affect liver [¹⁸F]FDG kinetics due to arterial and portal vein (PV) input. The purpose of this study was to evaluate kinetic models of [¹⁸F]FDG metabolism under a wide range of dietary interventions taking into account variations in arterial (HA) and portal vein (PV) input.

Methods—The study consisted of three groups of rats maintained under different diet interventions: 12 h fasted, 24 h fasted and those fed with high fructose diet. [¹⁵O]H₂O PET imaging was used to characterize liver flow contribution from HA and PV to the liver's dual input function (DIF). [¹⁸F]FDG PET imaging was used to characterize liver metabolism. Differences in [¹⁸F]FDG kinetics in HA, PV and liver under different diet interventions were investigated. An arterial to PV Transfer Function (TF) was optimized in all three dietary states to noninvasively estimate PV activity. Finally, two compartment 3-parameter (2C3P), two compartment 4-parameter (2C4P), two compartment 5-parameter (2C5P), and three compartment 5-parameter (3C5P) models were evaluated and compared to describe the kinetics of [¹⁸F]FDG in the liver across diet interventions. Sensitivity of the compartmental models to ratios of HA to PV flow fractions was further investigated.

Results—Differences were found in HA and PV [¹⁸F]FDG kinetics across 12 h fasted, 24 h fasted and high fructose fed diet interventions. A two exponential TF model was able to estimate portal activity in all the three diet interventions. Statistical analysis suggests that a 2C3P model configuration was adequate to describe the kinetics of [¹⁸F]FDG in the liver under wide ranging dietary interventions. The net influx of [¹⁸F]FDG was lowest in the 12 h fasted group, followed by 24 h fasted group, and high fructose diet.

Conclusions—A TF was optimized to non-invasively estimate PV time activity curve in different dietary states. Several kinetic models were assessed and a 2C3P model was sufficient to describe [¹⁸F]FDG liver kinetics despite differences in HA and PV kinetics across wide ranging dietary interventions. The observations have broader implications for the quantification of liver metabolism in metabolic disorders and cancer, among others.

Keywords

Diet interventions; High fructose diet; Portal vein; Transfer function; Liver metabolism; Dual input function; Small animal positron emission tomography (PET); Compartmental modeling

1. Introduction

Deficiencies in liver function are central to many diseases including diabetes, non-fatty alcohol disease and malignancies such as hepatocellular carcinoma (HCC) to name a few. Similarly, the use of [^{18}F]FDG with PET has proven to be indispensable in initial staging, management and response to therapy. In the liver, in particular, [^{18}F] FDG PET has been used in decision making as prognostic predictor of cancer [1,2], detection and evaluation of metastases to the liver [3,4], assessment of response to therapy [5,6] or as a preoperative tool to evaluate surgery risk [7–9]. While [^{18}F]FDG PET has been typically used to noninvasively quantify liver function in humans [10,11] and large animals [12–14], the contribution of the PV to liver kinetics is generally ignored. In addition, it is generally assumed that a 2-tissue compartment model is sufficient to describe the kinetics of [^{18}F]FDG in the liver. However, Southworth et al. [15] characterized the metabolism of [^{18}F]FDG in various tissues, including liver, using stable isotope techniques and reported that up to 45% of [^{18}F]FDG-6-phosphate (FDG6P) can be subject to further metabolism. To account for potential downstream metabolism of FDG6P in the liver, additional compartments and parameters may be needed.

In light of the above considerations, in this work, we performed small animal [^{18}F]FDG PET imaging to evaluate kinetic models of [^{18}F] FDG metabolism under a wide range of dietary conditions while taking into account both arterial and PV input to the liver. In particular, we accomplish the following aims: a) characterize differences in HA and PV [^{18}F]FDG kinetics and optimize a transfer function for the derivation of PV [^{18}F]FDG kinetics based on HA kinetics; b) reconstruct the DIF using [^{15}O]H₂O imaging to estimate respective flow fractions; and finally c) evaluate various models for [^{18}F]FDG kinetics in liver given the DIF. The above aims are performed under varying dietary interventions to characterize the dynamic range of results. Overall, the results enhance our understanding of [^{18}F]FDG metabolism in the liver across diet interventions to more accurately quantify liver function.

2. Materials and methods

All chemicals were purchased from Aldrich Chemical Co., Inc. Radioactive samples were counted on an 8000 γ -counter (Beckman Coulter). Small-animal PET was performed on either the microPET Inveon (Siemens Inc., Knoxville, TN) or Focus-220 [16] (Siemens Inc., Knoxville, TN). [^{15}O]-H₂O and [^{18}F]FDG were produced in the Washington University Cyclotron Facility.

2.1. Animal models and interventions

The study utilized male Zucker Lean (N=12) rats (Charles River Labs) at the age of 14 weeks. The rationale for the choice of animal model is primarily due to our experience with this animal model. In addition, the use of this model provides the basis for studying the involvement of the liver in diabetes and obesity by using the respective animal model littermates. In light of reports of downstream metabolism of [^{18}F]FDG in the liver [15], we sought to expand the dynamic range of [^{18}F]FDG metabolism and liver kinetics. To that end, the following interventions were considered: 1) 12 h fasted (12hF) (N=4) to promote low glucose utilization; 2) 24 h fasted (24hF) (N=4) to downgrade metabolic activity of glycogenesis; and 3) high fructose diet (HFrD) (N=4) comprising of 18.3% protein, 60.4%

carbohydrate and 5.2% fat by weight (Harlan Laboratories, Teklad Lab Animal Diets) for two weeks prior to imaging study to promote glycogenesis metabolic activity [17].

2.2. Animals preparation

For the fasted intervention groups, rats were fasted for 12 h or 24 h prior to imaging. The high fructose diet (HFrD) group was maintained on high fructose diet until time of imaging protocol. To anesthetize the rats prior to imaging, rats were placed in an induction chamber and allowed to breathe 1%–1.5% isoflurane (Butler Schein Animal Health) along with 1% oxygen until unconscious. During subsequent microsurgical procedures and imaging sessions, anesthesia was maintained via a nose-cone. Typically, anesthesia lasted for about 2 h. Body temperature was maintained by a circulating water blanket and a heat lamp. The PV of the rats was cannulated 1–2 weeks prior to imaging with a portal T-tube which was extended to allow for repeated blood sampling from the PV. In addition, a catheter was placed in femoral artery to obtain blood samples from the arterial line. No analgesia was used in these experiments. The rats were sacrificed following imaging per Washington University guidelines. All experiments were performed in compliance with the guidelines for the care and use of research animals established by Animal Studies Committee of Washington University School of Medicine in St. Louis.

2.3. Small animal PET protocol

Prior to administration of radiopharmaceuticals, each rat received a transmission scan to aid in attenuation correction. Following the transmission scan, dynamic acquisition was initiated 3 s before bolus injection of [^{18}F]FDG or [^{15}O]H₂O through the tail vein as described previously for multi-tracer small animal PET studies [18]. The injected dose for all the rats was about 18.5–37 MBq. For [^{18}F]FDG typically 10 to 15 blood samples (5 μL each) were obtained from arterial and portal lines during the 60 min dynamic acquisition, which were subsequently decay corrected to the time of injection. In addition, to estimate liver perfusion, a 10 min dynamic PET imaging with [^{15}O] H₂O was performed on the 12 h fasted rats (N=4) prior to administering [^{18}F]FDG. No blood sampling was performed during the [^{15}O]H₂O dynamic PET session.

2.4. Image processing

Image sinograms were reconstructed using filtered back projection with 40 frames for [^{18}F]FDG and 38 frames for [^{15}O]H₂O image data sets. A transmission scan preceding the emission scan allowed for attenuation correction, as mentioned above. To facilitate drawing of regions of interest (ROI), all the frames were summed into a single image and the ROIs were then drawn in the right lobe of the liver to obtain liver TAC. The ROIs from summed [^{18}F]FDG images were subsequently overlaid onto the corresponding [^{15}O]H₂O images to obtain the liver [^{15}O]H₂O kinetics.

2.5. Derivation of the input function

The arterial input function was reconstructed using the hybrid input blood sampling algorithm (HIBS) [19].

2.6. Optimization of HA-to-PV transfer function (TF)

Our approach is to optimize a Transfer Function (TF) describing the passage of tracer through the intestine into the portal vein [20,21]. The TF was represented by a sum of exponentials, with the PV activity described by the convolution of the TF with the arterial input function i.e.

$$\tilde{C}_{pv}(t) = [C_{HA}(t) \otimes h_T(t)] \quad (1)$$

where $\tilde{C}_{pv}(t)$ denotes the estimated PV activity, C_{HA} denotes the arterial input function and $h_T(t)$ denotes the TF. Two variations of TF comprising of a single exponential and sum of two exponentials were evaluated. The TF represented by a sum of two-exponential was found to fit the PV in all dietary states.

$$h_T(t) = P_1 e^{-P_2 t} + P_3 e^{-P_4 t} \quad (2)$$

To estimate the four parameters P_1 , P_2 , P_3 , and P_4 , the TF was fitted against the measured PV sampling data using nonlinear least squares algorithm. This fitting procedure was repeated in all the three diet interventions.

2.7. Modeling of [^{15}O]H $_2\text{O}$ kinetics

To estimate the portal venous and arterial flow contributions to the liver, a multi-tracer imaging protocol was followed in the 12hF group by imaging with [^{15}O]H $_2\text{O}$ prior to [^{18}F]FDG protocol, as described in the Small Animal PET Protocol section. We adapted the compartment model (Fig. 1) of Slimani et al. [22] and applied it to dynamic [^{15}O]H $_2\text{O}$ PET data to obtain estimates of flow and flow fractions. The parameters $F_{HA}(\text{mL}/\text{min}/\text{mL})$ and $F_{PV}(\text{mL}/\text{min}/\text{mL})$ represent flow rates from HA and PV, respectively, while $K = (F_{HA} + F_{PV})/V_L$ represents the clearance constant from the liver compartment with V_L capturing the volume of distribution in the liver. The time dependent concentration $C_L(t)$ in the liver compartment is obtained by solving the analytical equation for [^{15}O]H $_2\text{O}$ kinetics in the liver

$$C_L(t) = [F_{HA} (C_a(t) \otimes e^{-F_a t}) + F_{PV} C_{PV}(t)] \otimes e^{-Kt} \quad (3)$$

where $C_{PV}(t)$ is the concentration in the portal vein compartment. A hepatic blood volume term V_0 was included in the analysis to account for vasculature contribution to tissue activity

$$\tilde{C}_{ROI} = (1 - V_0)C_L(t) + V_0 C_a(t) \quad (4)$$

where $\tilde{C}_{ROI}$ is the observed activity. To estimate the flow from HA and PV (i.e. F_{HA} and F_{PV}) to the liver, the observed tissue TAC (Eq. 4) was used to fit against the liver ROI tissue TAC from [^{15}O]H $_2\text{O}$ images using weighted nonlinear least squares algorithm.

2.8. Kinetic modeling of [^{18}F]FDG

Several kinetic models were evaluated to account for metabolism of [^{18}F]FDG in the liver. Specifically we evaluated 4 variations of the typical 2-compartment model (Fig. 2): A) a 3-parameter (2C3P) model depicting trapping of [^{18}F]FDG metabolites; B) a 4-parameter (2C4P) model depicting phosphorylation and dephosphorylation of [^{18}F]FDG and FDG6P, respectively; C) a 5-parameter (2C5P) model with an added rate-constant to represent downstream metabolism of FDG6P and a D) three tissue compartmental model with 5-parameters (3C5P) to account for trapping of downstream metabolites. As applicable, K_1 (mL/min/g) and k_2 (/min) represent the bi-directional transport of [^{18}F]FDG from/to blood to/from tissue, k_3 (/min) denotes the phosphorylation rate-constant, k_4 (/min) denotes the dephosphorylation rate-constant, and k_5 (/min) denotes the rate-constant of down-stream metabolism.

The analytical solution for $C_L(t)$ in the 3P and 4P models has been reported previously [23]. Similarly, solving the algebraic equations in Laplacian domain, the concentrations $C_1(t)$ and $C_2(t)$ for the 2C5P model are obtained by taking the inverse Laplace Transform of

$$C_1(S) = \left[\frac{S + k_4 + k_5}{S^2 + s(k_2 + k_3 + k_4 + k_5) + (k_2k_4 + k_2k_5 + k_3k_5)} \right] K_1 C_a(S) \quad (5)$$

$$C_2(S) = \left[\frac{k_3}{S + k_4 + k_5} \right] C_1(S) \quad (6)$$

Total activity in the liver $C_L(t) = C_1(t) + C_2(t)$ is thus provided by

$$C_L(t) = \frac{K_1}{\alpha_1 - \alpha_2} C_a \otimes \left[(k_3 + k_4 + k_5 + \alpha_1) e^{\alpha_1 t} - (k_3 + k_4 + k_5 + \alpha_2) e^{\alpha_2 t} \right] \quad (7)$$

where

$$\alpha_{1,2} = \frac{-(k_2 + k_3 + k_4 + k_5) \mp \sqrt{(k_2 + k_3 + k_4 + k_5)^2 - 4(k_2k_4 + k_2k_5 + k_3k_5)}}{2}$$

The 3C5P is an extension of the 2C5P model with an additional tissue compartment to account for downstream metabolism and trapping of metabolites.

The impulse response of each of the compartmental models was convolved with the DIF and optimized against the liver PET data for all dietary interventions using

$$C_L(t) = (1 - V_0) h_L \otimes C_{DIF}(t) + V_0 C_{DIF}(t) \quad (8)$$

where $h_L(t)$ denotes the impulse response for a given compartmental model. The DIF was determined as the weighted sum of fractional contributions from HA and PV, i.e., $f_{HA} = F_{HA} / (F_{HA} + F_{PV})$ and $f_{PV} = F_{PV} / (F_{HA} + F_{PV})$, respectively. The total input to the liver from both HA and PV is thus provided by

$$C_{DIF}(t) = f_{HA} C_a(t) + f_{PV} C_{PV}(t) \quad (9)$$

Finally, we also evaluated total [^{18}F]FDG uptake using a graphical Patlak approach [24,25] for all diet interventions by estimating the net influx parameter (K_i),

$$K_i = \frac{K_1 k_3}{k_2 + k_3} \quad (10)$$

2.9. Data optimization

The Levenberg–Marquardt algorithm was used to minimize the sum of squares. A strict convergence criterion of 10^{-6} was set for all the cases studied. Prior to fitting all data were normalized to Standard uptake value (SUV) by multiplying with the ratio of animal weight to injected dose, which aided in normalization of initial parameter estimates. When using the weighted least squares, the Weighted Residual Sum of Squares (WRSS)

$$WRSS = \sum_{i=1}^N w_i [\tilde{C}_{Model}(t) - C_{ROI}(t)]^2$$
 was used to fit against the measured data where w_i is the weights, $C_{Model}(t)$ is the total observed activity from the model, $C_{ROI}(t)$ is the liver TAC obtained by drawing ROI and N is the number of data points. The weights used were proportional to square root of ratio of scan duration to tissue ROI activity i.e.

$w_i \propto \sqrt{\Delta t_i / C(t_i)_{ROI}}$. The values for the rate constants were constrained to be greater than zero in all the cases.

2.10. Sensitivity analysis

The sensitivity of the rate constants K_1 (mL/min/g), k_2 (/min) and k_3 (/min) with the flow fractions f_{HA} and f_{PV} was analyzed by considering five different HA-PV flow ratios i.e. 0.15:0.85, 0.2:0.8, 0.25:0.75, 0.3:0.7, 0.35:0.65. To that end, the effects of HA-PV flow ratio on the mean values of K_1 (mL/min/g), k_2 (/min) and k_3 (/min) in the 12hF diet intervention were evaluated by fitting the 3P model for each HA:PV ratio fraction. The percent bias in the parameter estimates due to variations in flow fractions was estimated in each rat relative to estimates obtained with HA:PV=0.11:0.89, i.e., $\%bias = 100(k^{\{HA:PV\}} - k^{\{0.11:0.89\}}) / k^{\{0.11:0.89\}}$

2.11. Statistical analysis

All the results were expressed as Mean $\pm$ SEM. To test for significant differences between diet interventions, a student t-test was performed and a P-value less than 0.05 was considered significant. When comparing the three model configurations, the corrected Akaike Information Criteria (AICc) [26], the Schwarz Criteria [27] and nested F-Test [28] were evaluated to assess the statistically favorable model. Lower scores of AICc and SC were preferred for the best model. For the F-Test, a P-value less than 0.05 was used to reject the nested simpler model in favor of the complex model with more number of parameters. When comparing the 2C5P and 3C5P configurations, only the AICc and SC were used in model selection since the 2C5P model is not considered a nested model of the 3C5P model.

3. Results

3.1. Effects of dietary interventions on arterial, PV and liver [18 F]FDG kinetics

The effects of dietary interventions on blood and liver [18 F]FDG kinetics are summarized in Fig. 3. On average, there is greatest concentration of [18 F]FDG in the arterial line after a 24hF followed by a HFrD and then 12hF intervention (Fig. 3A). On average significant variations in the PV kinetics of [18 F]FDG were observed between the three diet interventions (Fig. 3B) with PV [18 F]FDG kinetics in 12hF group being significantly higher in comparison to 24hF and HFrD groups. On average we did not observe differences in the kinetics of [18 F]FDG in the liver under the dietary interventions tested (Fig. 3C).

3.2. HA-to-PV transfer function (TF)

Having characterized arterial-PV [18 F]FDG kinetics, we proceeded to optimize a TF to estimate PV kinetics based on arterial kinetic data. The estimated parameter values for the TF in all the three diet interventions are summarized in Table 1. Although average parameter estimates were not statistically significant between dietary interventions, we observed trends in estimated values between groups. For example P_1 was about two-fold higher in 12hF when compared to 24hF and HFrD groups while P_0 was zero in the 12hF group and nonzero is 24hF and HFrD groups. A representative fit for each intervention is displayed in Fig. 4.

3.3. Estimation of HA and PV flow fractions

We fitted [^{15}O]H $_2$ O kinetic data to the liver perfusion model using 12 h fasted rats to estimate the hepatic perfusion parameters. The values for parameters F_{HA} , F_{PV} , v_0 and corresponding fractional contribution F_{HA} and F_{PV} are summarized in Table 2. The mean values obtained for F_{HA} and F_{PV} were 0.43 ± 0.09 and 3.28 ± 0.24 mL/min/mL, respectively, resulting in mean arterial and portal flow fractions $f_{HA} = 0.11 \pm 0.02$ and $f_{PV} = 0.89 \pm 0.02$, respectively. In subsequent analysis of [^{18}F]FDG kinetics, we use above-mentioned flow fraction to optimize kinetic estimates.

3.4. Compartmental modeling of [^{18}F]FDG kinetics in the liver

The parameter estimates for the four model configurations are summarized in Table 3 for the various diet interventions. The estimates were evaluated with the DIF which was obtained by using flow fractions of $f_{HA}=0.11$ and $f_{PV}=0.89$. In all the diet interventions, K_1 , k_2 and k_3 were on an average higher in 2C4P and 2C5P than 2C3P models. However, based on the statistical analysis (i.e. AICc, SC and F-test), the 2C3P model was favored in all studies compared to the 2C4P model except in one study in the 24hF group where a 2C4P model was favored. In addition, the 2C5P and 3C5P models offered no significant advantage over 2C3P or 2C4P model configurations. For this reason, all subsequent analysis utilized the 2C3P model.

Typical fits are shown in Fig. 5 for each intervention. In the 12hF group, the average K_1 was significantly lower than the average K_1 of the 24hF and HFrD groups. On average, the reverse rate constant k_2 (/min) in the 12hF group was significantly lower ($P < 0.01$) than 24hF group. The phosphorylation rate-constant k_3 (/min) of 12hF group was approximately 3-fold lower in comparison to the 24hF group and 4-fold ($P < 0.001$) lower in comparison to the HFrD group. The net influx constant K_i in the 12hF group was lower, though marginally not significant, in comparison to 24hF group and significantly lower in comparison to the HFrD group (Fig. 5D).

3.5. Sensitivity of 3P model to HA:PV flow fractions

The sensitivity of the 3P model to variations in HA:PV flow fractions was evaluated. The percentage bias in estimates of K_1 , k_2 , k_3 and K_i at each HA:PV flow fraction ratio relative to estimates with $f_{HA}:f_{PV}=0.11:0.89$ for each study and the average bias are shown in Fig. 6. In general, with increasing $f_{HA}:f_{PV}$ ratio, there is a decrease in parameter estimates. Overall, the rate-constants K_1 and k_2 were most sensitive to variations in HA:PV flow fractions in the 12hF group.

4. Discussion

Quantification of liver function is hindered by the dual input nature from both the hepatic artery and portal vein to the liver with the latter being inaccessible. While some work to estimate to the PV has been performed in humans [11,12,29] and in large animals [20,21], none has been performed in rodents. To that end, in the current work, we characterized the kinetics of [^{18}F]FDG in HA, PV and liver following 12hF, 24hF and HFrD interventions to expand the dynamic range of liver glucose metabolism. To facilitate reconstruction of the liver dual input function, we optimized a TF for the prediction of PV kinetics based on arterial kinetics under wide ranging dietary interventions. The flow fraction contribution to the input function was estimated from kinetic analysis of [^{15}O]H $_2$ O images. Finally, four compartmental models were evaluated for the kinetics of [^{18}F]FDG in the liver under the three dietary interventions.

Consideration of the fasting state and dietary intervention is critical for accurate quantification of tissue function. Previous studies documented the effects of fasting and anesthesia on arterial and radiopharmaceutical tissue kinetics [30–33]. However, to our knowledge, this work is the first to highlight differences in PV kinetics under various dietary conditions to facilitate quantitative imaging of the liver. The results indicate differences in arterial and PV [^{18}F]FDG kinetics, thus highlighting the need to account for PV contribution in quantification of liver function under various dietary conditions. For example, [^{18}F]FDG - PET has been used in assessing allograft rejection in a rat liver model of liver transplantation [34], hepatic cancer imaging [35], as well as in derivation of the arterial input function in mice [36]. More recently [^{18}F]FDG -PET has been used to investigate the role of blood flow and intrinsic tissue clearance in obese and type 2 diabetic rats [37]. In all such studies, the contribution of the PV to liver input was neglected in pre-clinical studies, thus limiting insight or biasing reported results. Interestingly, while the dietary interventions resulted in alteration of HA and PV kinetics, liver kinetics under all dietary interventions was not significantly different (Fig. 3C) as discussed further below.

Having characterized arterial-PV kinetics across various diet interventions, we optimized a TF to predict PV [^{18}F]FDG kinetics. The optimized TF was inspired by previous work on optimization of arterial-to-PV TF in large animals [20,21]. The TF was able to fit the PV in all the studies. The optimized parameter P_4 was zero in 12hF and nonzero in 24hF and HFrD groups, highlighting the role of substrate metabolism in the gut and the contribution to total liver input. Individual variations in parameters estimates under each intervention further suggest that the TF is dependent on the conditions of the study. The estimated portal activity was then used to reconstruct the DIF in different diet interventions with mean flow fractions of $f_{HA} = 0.11$ and $f_{PV} = 0.89$ obtained from the [^{15}O]H $_2$ O imaging studies. The mean arterial and portal flows F_{HA} and F_{PV} were 0.43 ± 0.09 and 3.28 ± 0.04 mL/min/mL and are in agreement with previously reported total hepatic blood flow values in perfused rat livers [38]. In the 24hF and HFrD groups, the mean arterial and PV flow fractions from the 12hF group were used to reconstruct the DIF. While variations in blood flow through the PV are not expected to change between 12hF and 24hF, in the fed state blood flow may be increased through the gut system due to hepatic circulation regulatory mechanisms [37,39]. In addition, sensitivity analysis was performed to account for the effect of varying arterial and portal flow fractions in the 12hF group.

In light of a report suggesting presence of metabolites beyond FDG6P in the liver [15] we evaluated four models for [^{18}F]FDG kinetics in the liver: the classic 2C3P model; a 2C4P model with the fourth parameter (k_4) denoting potential activity of glucose-6-phosphatase; a 2C5P model with a fifth lumped-parameter (k_5) denoting downstream metabolism of [^{18}F]FDG, and a 3C5P model with k_5 representing downstream metabolism and trapping of metabolites. Since the enzyme glucose-6-phosphatase is known to be present in the liver [15], in contrast to brain and heart, we anticipated non-zero k_4 parameter estimates. On the contrary, in most cases k_4 was found to be negligible (or zero) across all dietary interventions. This observation suggests that most of the activity is in the form of [^{18}F]FDG, as observed previously, and is likely due to the high K_m of the liver's type IV hexokinase limiting the phosphorylation of [^{18}F]FDG [15]. In addition, k_5 was found to be zero (or negligible) under most conditions for both the 2C5P and 3C5P model configurations suggesting negligible downstream metabolism and trapping. Visual inspection of the normalized liver TAC suggests that the kinetics of [^{18}F]FDG in the liver is essentially identical despite wide ranging dietary interventions. Statistical analysis on the model subsets supports the visual observations in that the 3P model was sufficient to explain [^{18}F]FDG kinetics in the liver for all interventions.

While in general parameter values are consistent within a given model configuration, there is some variability in both the 12hF and 24hF groups, in particular in reference to animal #4. Although temperature was maintained constant, we did not collect other physiologic parameters that may explain the variability. However, while variable, the main thrust of the work is to characterize the appropriate kinetic model to account for FDG metabolism in the liver. From this point of view, the optimal kinetic model is consistently the 2C3P model configuration. Additionally, due to the low blood volume of rats and the multiple sampling (arterial, portal vein) and time points, we could not determine the plasma-to-blood partition ratio of FDG for individual animals, which may affect the input function as only FDG in plasma is usually assumed to be available to transport into tissue. In quantification of heart and liver FDG kinetics, both human and animal, the partition ratio has generally been assumed to be constant with the underlying assumption that the equilibrium between plasma and whole blood is rapid [11,40–43]. In either of the abovementioned scenarios, the partition ratio will not affect the overall interpretation of the results since the ratio would be a constant scaling applied to all input functions. However, a nonlinear partition ratio may affect the input function, although we anticipate that any non-linearity in the partition ratio, will affect early kinetics, i.e., the uptake phase of the kinetics.

The compartmental analysis using the 3P model indicated a higher overall transport rate-constant of [^{18}F]FDG in the 24hF and HFrD groups when compared to the 12hF group. A similar increase in hepatic glucose transport was found after oral loading in rats [39]. In addition, the 4-fold higher phosphorylation rate-constant (k_3) in the HFrD group in comparison to the 12hF group is in agreement with reported increase in k_3 after fructose administration [44]. Interestingly, [^{18}F]FDG transport and phosphorylation rate-constants increased in the 24hF intervention group in comparison to the 12hF group. Overall, these observations are also evident when examining the total glucose uptake constant (K_i). Following an extended fast, tissue uptake of glucose is generally inhibited to support glucose needs of critical tissues such as brain and heart. In the liver, in particular, GLUT2 gene and protein expression may be modulated to that end. In addition, glycogen stores in the muscle and liver undergo glycogenolysis to raise peripheral glucose concentrations [45]. Thus, the observation that glucose transport is increased following a 24hF is surprising, yet may indicate a transient uptake of glucose [46]. Finally, sensitivity analysis of kinetic estimates to variations in HA:PV flow fractions in the 12hF suggests a decrease in estimates with increasing flow fraction ratio, particularly with K_1 and k_2 being most sensitive.

5. Conclusions

In the current work, we have characterized arterial and PV [^{18}F] FDG differences across various diet interventions and evaluated four different kinetic models to facilitate quantification of [^{18}F]FDG metabolism in the liver. Our data indicate that arterial-PV [^{18}F]FDG kinetics varied across diet interventions. Statistical analysis of [^{18}F] FDG compartmental models suggests that a 2-compartment 3P model is sufficient to describe the kinetics of [^{18}F]FDG in the liver. Further studies are needed to characterize quantification of metabolic tracers, including C-11 radiopharmaceuticals, in quantifying liver substrate metabolism in disease states such as diabetes, obesity among others.

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Fig. 1.

Kinetic model used to estimate the flow fractions using $[^{15}\text{O}]\text{H}_2\text{O}$ as described previously [22] where F_{HA} and F_{PV} denote the HA and PV flow rates, respectively; $C_D(t)$ denotes a dispersion compartment; $C_{\text{PV}}(t)$ and $C_{\text{GI}}(t)$ represent the PV and GI compartments, respectively; and $C_L(t)$ denotes the activity in the liver. The clearance rate constant from the liver is denoted by $K = (F_{\text{HA}} + F_{\text{PV}})/V_L$.

Fig. 2.

Kinetic models for hepatic [^{18}F]FDG metabolism comprising of two-tissue compartments with A) 3 Parameters (2C3P) B) 4 Parameters (2C4P) C) 5 Parameters (2C5P) D) three compartmental model with 5 Parameters (3C5P). Depending on the model, K_1 (mL/min/g) and k_2 (/min) represent the bi-directional transport of [^{18}F]FDG from/to blood to/from tissue, k_3 (/min) denotes the phosphorylation rate-constant, k_4 (/min) denotes the de-phosphorylation rate-constant, and k_5 (/min) denotes the rate-constant of down-stream metabolism.

Fig. 3.

Top row: comparison of normalized kinetics of $[^{18}\text{F}]\text{FDG}$ (SUV) in (A) arterial, (B) PV, and (C) liver across various diet interventions. Bottom row: Comparison of arterial vs. PV $[^{18}\text{F}]\text{FDG}$ kinetics following D) 12hF (N=4) E) 24hF (N=4) F) HFrD (N=4) for 2 weeks. *denotes 12hF significantly higher than 24hF group and #denotes 12hF significantly higher than HFrD group. Values represent mean $\pm$ SEM.

Fig. 4. Representative fits in optimizing a 2-exponential TF to PV $[^{18}\text{F}]\text{FDG}$ kinetics in the A) 12hF group B) 24hF group C) HFrD group per Eqs. 1–2 of the Methods Section.

Fig. 5. Representative fits of the two-tissue 3P compartmental model of Fig. 2A to liver data using the reconstructed DIF in the A) 12hF group B) 24hF group C) HFrD group D) Variation in the glucose influx constant K_1 (mean $\pm$ SEM) across the three diet interventions (N=4 per group).

Fig. 6. Percent (%) bias in A) mean K_1 (mL/min/g) B) mean k_2 (/min) and C) mean k_3 (/min) D) mean K_i (mL/min/g) to the HA-to-PV flow fraction ratio (HA:PV) relative to HA:PV= 0.11:0.89 computed using a two tissue 3P model in the 12hF group (N=4).

Table 1

Parameter estimates for TF across various diet interventions.

Rat #	P ₁	P ₂ (min ⁻¹)	P ₃	P ₄ (min ⁻¹)
12hF				
1	4.56	1.30	-0.0160	0.0000
2	1.98	0.92	-0.0121	0.0000
3	4.28	1.80	-0.0099	0.0000
4	2.03	0.92	-0.0065	0.0000
Avg.	3.21 ± 0.69	1.23 ± 0.21	-0.0111 ± 0.0019	0.0000 ± 0.0000
24hF				
1	0.99	0.78	-0.0096	0.0001
2	2.79	1.16	-0.1104	0.0692
3	1.53	1.13	-0.0214	0.0171
4	0.92	0.76	-0.0097	0.0183
Avg.	1.56 ± 0.43	0.96 ± 0.11	-0.0378 ± 0.0244	0.0262 ± 0.0149
HFrD				
1	0.89	0.70	-0.0106	0.0145
2	1.63	1.02	-0.6212	0.8840
3	1.15	1.80	0.2219	0.5562
4	2.40	2.85	0.0222	0.1465
Avg.	1.52 ± 0.33	1.59 ± 0.48	-0.0969 ± 0.1822	0.4003 ± 0.1982

Data shown are mean ± SEM.

Table 2

Estimated HA and PV flow fractions using [¹⁵O]H₂O imaging studies group.

Rat no	F _{HA} (mL/min/mL)	F _{PV} (mL/min/mL)	f _{HA}	f _{PV}	V ₀
1	0.57	3.95	0.13	0.87	0.02
2	0.37	2.84	0.11	0.89	0.05
3	0.57	3.25	0.15	0.85	0.01
4	0.20	3.09	0.06	0.94	0.02
Average	0.43 ± 0.09	3.28 ± 0.24	0.11 ± 0.02	0.89 ± 0.02	0.03 ± 0.01

Data shown are mean ± SEM.

Table 3

Comparison of models for various diet interventions with the F-Test denoting the preferred model between the three models.

A. Two tissue 3-parameter compartmental model						
3P						
#	K ₁	k ₂	k ₃	K ₁	AICc	SC
12hF						
1	2.14	5.21	0.0024	0.0010	18.8	23.8
2	3.23	5.76	0.0032	0.0018	-24.2	-19.2
3	1.43	1.67	0.0024	0.0021	7.0	12.0
4	2.28	3.07	0.0005	0.0004	55.1	60.1
Avg.	2.27 ± 0.37	3.93 ± 0.95	0.0021 ± 0.0005	0.0013 ± 0.0004		
24hF						
1	12.23	10.84	0.0052	0.0059	52.7	57.6
2	7.68	10.14	0.0099	0.0075	-12.3	-7.3
3	8.07	7.74	0.0071	0.0072	4.0	9.0
4	13.97	12.00	0.0003	0.0004	78.4	83.3
Avg.	10.49 ± 1.55 ^a	10.18 ± 0.90 ^a	0.0056 ± 0.0020	0.0052 ± 0.0016 [*]		
HFD						
1	8.38	5.71	0.0083	0.0122	25.0	30.0
2	11.02	12.15	0.0079	0.0072	49.2	54.2
3	6.58	6.50	0.0096	0.0097	52.8	57.8
4	4.79	4.95	0.0082	0.0079	48.4	53.4
Avg.	7.69 ± 1.33 ^a	7.33 ± 1.64	0.0085 ± 0.0037 ^a	0.0092 ± 0.0011 ^a		
B. Two compartment 4-parameter (2C4P) model						
4P						
#	K ₁	k ₂	k ₃	k ₄	AICc	SC F-Test
12hF						
1	2.12	5.26	0.0024	0.0000	21.5	27.3 2C3P
2	3.17	5.66	0.0033	0.0000	-21.0	-15.1 2C3P
3	1.66	1.93	0.0024	0.0000	13.1	19.0 2C3P

B. Two compartment 4-parameter (2C4P) model						
4P						
#	K ₁	k ₂	k ₃	k ₄	AICc	SC F-Test
4	2.28	3.12	0.0025	0.0537	58.6	64.5 2C3P
Avg:	2.31 ± 0.32	3.99 ± 0.89	0.0027 ± 0.0002	0.0134 ± 0.1334		
24hF						
1	11.94	10.56	0.0053	0.0000	55.4	61.3 2C3P
2	8.56	12.28	0.0206	0.0220	-17.8	-11.9 2C4P
3	8.01	7.68	0.0069	0.0000	6.8	12.7 2C3P
4	13.89	11.93	0.0003	0.0000	81.2	87.1 2C3P
Avg:	10.60 ± 1.40 ^b	10.61 ± 1.05 ^b	0.0083 ± 0.0043	0.0055 ± 0.0055		
HFrd						
1	8.79	5.98	0.0083	0.0000	27.2	33.1 2C3P
2	11.14	12.34	0.0083	0.0020	52.5	58.4 2C3P
3	6.85	7.18	0.0169	0.0170	63.3	69.1 2C3P
4	4.81	5.20	0.0134	0.0151	56.4	62.3 2C3P
Avg:	7.90 ± 1.35 ^b	7.67 ± 1.61	0.0117 ± 0.0021 ^b	0.0085 ± 0.0044		

C. Two compartment 5-parameter (2C5P) model kinetic estimates						
5P						
#	K ₁	k ₂	k ₃	k ₄	k ₅	AICc SC F-Test
12hF						
1	2.09	5.19	0.0024	0.0000	0.0000	24.6 31.2 2C4P
2	3.28	5.84	0.0032	0.0000	0.0000	-18.6 -11.9 2C4P
3	1.45	1.69	0.0023	0.0000	0.0000	13.6 20.2 2C4P
4	2.28	3.12	0.0025	0.0242	0.0293	61.6 68.2 2C4P
Avg:	2.27 ± 0.37	3.96 ± 0.95	0.0026 ± 0.0002	0.0061 ± 0.0061	0.0073 ± 0.0073	
24hF						
1	12.12	10.67	0.0055	0.0000	0.0000	58.3 65.0 2C4P
2	8.56	12.28	0.0206	0.0117	0.0102	-14.8 -8.2 2C4P
3	7.97	7.65	0.0071	0.0000	0.0000	9.7 16.4 2C4P
4	13.66	11.73	0.0003	0.0000	0.0000	84.1 90.8 2C4P
Avg:	10.58 ± 1.38 ^c	10.58 ± 1.03 ^c	0.0084 ± 0.0043	0.0029 ± 0.0029	0.0026 ± 0.0026	

C: Two compartment 5-parameter (2C5P) model kinetic estimates							
5P							
#	K ₁	k ₂	k ₃	k ₄	k ₅	AICc	SC F-Test
HFrd							
1	8.42	5.73	0.0083	0.0000	0.0000	30.4	37.1 2C4P
2	11.13	12.33	0.0084	0.0000	0.0020	55.5	62.1 2C4P
3	6.88	7.22	0.0169	0.0169	0.0000	66.2	72.9 2C4P
4	4.78	5.16	0.0134	0.0149	0.0001	59.4	66.0 2C4P
Avg.	7.80 ± 1.34 ^c	7.61 ± 1.63	0.0118 ± 0.0021 ^c	0.0080 ± 0.0046	0.0005 ± 0.0005		
D: Three compartment 5-parameter (3C5P) model kinetic estimates							
5P							
#	K ₁	k ₂	k ₃	k ₄	k ₅	AICc	SC F-Test
12hF							
1	2.18	5.39	0.0024	0.0001	0.0000	23.63	30.3 N/A
2	2.82	5.10	0.0033	0.0000	0.0000	-15.16	-8.5 N/A
3	1.42	1.65	0.0020	0.0000	0.0000	17.78	24.5 N/A
4	2.28	3.23	0.0025	0.0535	0.0000	60.42	67.1 N/A
Avg.	2.17 ± 0.29	3.84 ± 0.82	0.0025 ± 0.0002	0.0134 ± 0.0061	0.0000 ± 0.0000		
24hF							
1	12.74	11.34	0.0050	0.0000	0.0000	56.87	63.5 N/A
2	8.37	12.18	0.0196	0.0206	0.0018	-24.18	-17.5 N/A
3	8.71	9.09	0.0069	0.0000	0.0120	3.00	9.63 N/A
4	13.88	11.81	0.0003	0.0000	0.0000	82.8	89.4 N/A
Avg.	10.92 ± 1.39 ^d	11.10 ± 0.69 ^d	0.0080 ± 0.0041	0.0052 ± 0.0051	0.0034 ± 0.0028		
HFrd							
1	8.45	5.75	0.0083	0.0000	0.0000	30.36	37.0 N/A
2	11.09	12.31	0.0085	0.0020	0.0000	55.33	61.9 N/A
3	6.86	7.19	0.0169	0.0169	0.0000	66.30	72.9 N/A
4	4.79	5.18	0.0134	0.0151	0.0000	59.36	66.0 N/A
Avg.	7.79 ± 1.32 ^d	7.60 ± 1.62	0.0118 ± 0.0021 ^d	0.0085 ± 0.0043	0.0000 ± 0.0000		

The 3P, 4P and 5P models indicate a (A) two tissue 3-parameter, (B) two tissue 4-parameter (C) two tissue 5-parameter and (D) three tissue 5-parameter models.

* Denotes marginally not significant at $P=0.06$. Data shown are mean $\pm$ SEM.

^a Denotes significantly higher than 12hF. Data shown are mean $\pm$ SEM.

^b Denotes significantly higher than 12hF; F-Test comparison 4P vs 3P. Data shown are mean $\pm$ SEM.

^c Denotes significantly higher than 12hF; F-Test comparison 5P vs 4P. Data shown are mean $\pm$ SEM.

^d Denotes significantly higher than 12hF; F-Test comparison 5P vs 3P. Data shown are mean $\pm$ SEM; N/A, not applicable.