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Oxygen transport in a cross section of the rat inner medulla: Impact of heterogeneous distribution of nephrons and vessels

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Abstract

We have developed a highly detailed mathematical model of oxygen transport in a cross section of the upper inner medulla of the rat kidney. The model is used to study the impact of the structured organization of nephrons and vessels revealed in anatomic studies, in which descending vasa recta are found to lie distant from clusters of collecting ducts. Specifically, we formulated a two-dimensional oxygen transport model, in which the positions and physical dimensions of renal tubules and vessels are based on an image obtained by immunochemical techniques (Pannabecker and Dantzer, *Am J Physiol Renal Physiol*, 2006). The model represents oxygen diffusion through interstitium and other renal structures, oxygen consumption by the Na^+/K^+ -ATPase activities of the collecting ducts, and basal metabolic consumption. Model simulations yield marked variations in interstitial PO_2 , which can be attributed, in large part, to the heterogeneities in the position and physical dimensions of the collecting ducts. Further, results of a sensitivity study suggest that medullary oxygenation is highly sensitive to medullary blood flow, and that, at high active consumption rates, localized patches of tissue may be vulnerable to hypoxic injury.

Keywords

mathematical model; hypoxia; rat kidney; inner medulla; vasculature

1. Introduction

The proper matching of oxygen supply to demand in a variety of tissues is necessary to preserve tissue viability. In the mammalian kidneys, despite high blood flow and oxygen delivery, oxygen tensions are relatively low, especially in the renal medulla, which have been measured to be ~ 20 and 10 mmHg in the outer and inner medulla, respectively [1]. (The medulla of a mammalian kidney can be divided into two sections: outer medulla and inner medulla.) Thus, the kidney exhibits a marked discrepancy between blood supply and oxygenation. One reason for this is that despite high overall blood flow to the kidney, the fraction of that flow reaching the medulla is relatively low, especially in the inner medulla (which receives $< 1\%$ of total renal blood flow [2]). In addition to reduced medullary O_2

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supply, low renal oxygen tension can be attributed, in large part, to the kidney's high oxygen consumption rate per weight of tissue, which is second only to the retina in the eye [3].

Blood vessels that perfuse the renal medulla of the rat kidney, which is one of the most well-studied kidneys, arise from the efferent arterioles of mid-cortical and juxtamedullary glomeruli. After entering the outer medulla, efferent arterioles branch into descending vasa recta (DVR). The DVR reach into differing depths of the medulla, then break into capillaries that perfuse that region of the medulla.

Within the medulla, the nephron segments that have the highest metabolic demands are the thick ascending limbs, which are found in the outer medulla. The Na^+/K^+ -ATPase of the thick ascending limbs consumes the greatest portion of oxygen in the renal medulla. The combination of high oxygen demand and low oxygen tension results in the thick ascending limb cells operating at high risk of hypoxic injuries, on one hand, and limits oxygen delivery to the inner medulla, on the other.

The availability of oxygen to a given nephron segment depends not only on the supply-and-demand ratio, but also on the distance between that nephron segment and the oxygen-supplying vessels. Anatomic studies in the medulla of rodent kidneys have revealed a highly structured organization of nephrons and vessels. That structural organization can be found in the outer medulla and in the upper inner medulla. The spatial arrangement of tubules and vessels in the outer medulla has been known for decades [4, 5, 6, 7], and the resulting impact on solute transport and medullary oxygenation has been considered in a number of studies [8, 9, 10, 11].

In contrast, solute transport in the inner medulla is much less well understood. Thus, the present study will focus on the organization of tubules and vessels in the upper inner medulla. Anatomic studies by Pannabecker and co-workers revealed clusters of collecting ducts (CD) in the upper 3–3.5 mm of the inner medulla [12]. The oxygen-supplying descending vasa recta are found distant from the collecting duct clusters. Note that unlike the thick ascending limbs in the outer medulla, the thin ascending limbs, which are the inner medullary ascending segments of loops of Henle that reach into the inner medulla, do not engage in significant active transport [13]. In contrast, functional and modeling studies suggest a brisk active NaCl reabsorption in the collecting duct [14, 15]. Given these considerations, we are motivated to study functional implications of the spatial arrangement of tubules and vessels on oxygenation in the upper inner medulla.

Chen et al. have used a mathematical model of solute transport to study the impact of structural organization in the outer medulla of the rat kidney on oxygenation [8, 9]. Their simulation results suggest that the sequestration of the DVR from the thick ascending limbs yields a substantial radial gradient (perpendicular to the corticomedullary axis) in oxygen tension. The model by Chen et al. [8, 9] represents only the outer medulla; the inner medulla is not included.

To better understand oxygenation in the inner medulla, we have developed a mathematical model of oxygen transport in a cross section of the upper inner medulla of the rat kidney. The distribution of tubules and vessels is generally believed to be more homogeneous in the

deep inner medulla; thus we focus on the upper inner medulla. The position and physical dimensions of the tubules are based on an image by Pannabecker and Dantzler [12], taken 400 μm below the outer-inner medullary boundary. Now, even with the heterogeneous distribution of the medullary structures, it is not immediately clear that there will be noticeable variations in oxygen tension at a given medullary level. This is because the tubules and the collecting duct clusters are rather small, with the mean separation between a collecting duct and DVR less than 100 μm .

Thus, the principal goal of this study is to assess the extent to which the exclusion of the DVR from the collecting duct clusters can generate a substantial radial gradient in oxygen tension. In addition, we will investigate whether this heterogeneous spatial arrangement of inner medullary tubules and vessels potentially exposes the kidney to regions of low tissue oxygen tension, where renal hypoxic injury is likely to occur.

2. Methods

2.1. Inner medullary architecture

The model was constructed with the positions and physical dimensions of the tubules and vessels based on an image by Pannabecker and Dantzler [12], taken 400 μm below the outer-inner medullary boundary of the rat kidney, and shows positions of collecting ducts, DVR, and ascending vasa recta (AVR). The image is reproduced in Fig. 1. The size of the simulation domain was $204 \times 204 \mu\text{m}$, to correspond with the image size. Within this image, there were 19 DVR, 84 AVR, and 15 collecting ducts. From the data set, it was determined that the average cross-sectional areas of the inner medullary DVR, AVR, and collecting duct were 42.4, 26.0, and 244 μm^2 , respectively. For simplicity, it was assumed that these cross-sectional areas could be well-approximated by a circular disk with that area.

As previously noted, a goal of this study is to assess the impact of the heterogeneous spatial distribution of tubules and vessels on medullary oxygenation. Thus, we investigated two scenarios, based on the number of tubules and vessels in the image by Pannabecker and Dantzler. In the first, these vasa recta and collecting ducts were distributed uniformly (equally) across the tissue cross section; in the second, they were distributed in the actual spatial locations determined by the Pannabecker and Dantzler's images. Figures 2a and 2b show these two different vessel and tubule distributions.

In addition to the heterogeneity in spatial location indicated by Pannabecker and Dantzler's image, there was also a noticeable spread in the size of the collecting ducts. Collecting duct luminal diameter affects the total rate of active NaCl transport, and thus likely has a significant impact on oxygen distribution. Given this observation, two more architectures were created that incorporated the experimental range of collecting duct diameters, to examine what effect this heterogeneity in size had as well (described further below). This gave us four cases to consider (with abbreviations): (1) Equally-spaced tubules/vessels, Same-sized collecting ducts (ES); (2) Equally-spaced tubules/vessels, Heterogeneous collecting duct sizes (EH); (3) Actual-spaced tubules/vessels, Same-sized collecting ducts (AS); (4) Actual-spaced tubules/vessels, Heterogeneous collecting duct sizes (AH).

The degree to which results from this study can be more generally applied to the upper inner medulla depends upon how representative the cross-sectional image used here is of the distribution of tubules and vessels across a range of images. Larger transverse images from Ref. [12] (see Fig. 1 from that study) showing distributions of DVR and AVR (and, separately, CDs) demonstrate similar patterning (clustering of CDs) to that from Figure 1. Further, a more quantitative study using multiple rat kidneys from Pannabecker et al. in 2008 [16] demonstrated that DVR do lie significantly away from CD clusters at the cluster boundaries. In addition, Pannabecker and Dantzler [12] found that the mean number of AVR surrounding CDs for five different kidneys was 4.12 ± 0.15 , compared to 4 for our particular image. Thus, while there may be some variability in exact features, the heterogeneous structure attempting to be captured by this study seems to be fairly representative of that present throughout the upper inner medulla.

2.2. Oxygen transport model

The dissolved oxygen concentration in tissue is proportional to the partial pressure of oxygen, or PO_2 , where the proportionality constant α is the solubility of oxygen in tissue. The distribution of tissue PO_2 was determined using a two-dimensional diffusion equation with consumption and production:

$$\alpha \frac{\partial}{\partial t} [P(x, y)] = \alpha \nabla \cdot [D(x, y) \nabla P(x, y)] - M(P, x, y) + S_C, \quad (1)$$

where $P(x, y)$ is the tissue PO_2 , $D(x, y)$ is the diffusion coefficient in the tissue, $M(P, x, y)$ is the rate of oxygen consumption in tissue (which is assumed to be a function of the PO_2 , as well as location within the cross section), and S_C is an oxygen source term due to capillaries and tubules not captured in the image from [12]. Periodicity was assumed at the computational domain boundaries. The tissue cross section was assumed to be homogeneous with respect to O_2 diffusion, so that the coefficient D is set to a fixed value of $1.28 \times 10^{-5} \text{ cm}^2/\text{s}$ and the solubility α is set to a fixed value of $4.24 \times 10^{-5} \text{ mL } O_2 \text{ mL}^{-1} \text{ mmHg}^{-1}$ [17] at every tissue point. The rate of oxygen consumption was assumed to be the sum of basal tissue oxygen consumption throughout the cross section, M_B , and active tissue oxygen consumption near the collecting duct wall, M_A , due to active transport of solutes. That is, $M = M_A + M_B$.

M_B was assumed to behave with Michaelis-Menten-like kinetics, with

$$M_B(P, x, y) = M_0 \frac{P}{P_0 + P}, \quad (2)$$

where M_0 is the maximal rate of oxygen consumption when oxygen is not limiting, and P_0 is the PO_2 at which the consumption is half-maximal. Here, M_0 and P_0 were assumed constant throughout the tissue with values of $10 \text{ } \mu\text{M/s}$ [9] and 10 mmHg [18, 3], respectively.

As previously noted, the inner-medullary segments of the loops of Henle are not known to engage in significant active solute transport, and neither are the vasa recta. In contrast, the inner medullary collecting ducts are believed to engage in brisk active transport of NaCl [14, 15]. The extent to which the collecting duct transport is supported by aerobic versus

anaerobic metabolism is controversial [19, 20]; thus, M_A was a value that was varied. *In vivo*, M_A depends, in large part, on the luminal concentration of Na^+ , which was assumed known, since the model only tracks tissue PO_2 in the cross section.

We assumed that the active transport pumps are distributed at a uniform density along the collecting duct wall, so that the rate of active transport is larger in tubules with larger radii. Thus, the rate of oxygen consumed near a collecting duct wall was represented by the product of a baseline active consumption rate and the relative radius of the collecting duct. In the images by Pannabecker and Dantzler, not all collecting ducts have the same radius. The differences in radius have the largest impact on consumption of oxygen; thus, these differences were represented as differences in M_A . Although the precise value of M_A is not well understood, previous modeling studies have indicated that around 97% of oxygen consumption in the medulla is due to active processes [8, 9]. Thus, for tissue points at the collecting duct wall interface, the baseline active transport rate was calculated as

$M_A = \frac{97}{3} M_0 \frac{R}{R_{avg}}$ where R is the radius of the adjacent collecting duct, and R_{avg} is the average radius of the collecting ducts in the simulation. The factor $\frac{97}{3}$ was chosen so that for a collecting duct with average radius $R = R_{avg}$, total oxygen consumption is given by

$$M(P, x, y) = \frac{97}{3} M_0 \frac{R_{avg}}{R_{avg}} + M_B(P, x, y). \quad (3)$$

When oxygen is not rate-limiting for basal consumption, $M_A = 97\%$ of M . For all other tissue points, $M_A = 0$.

Also present in the inner medulla but not captured in the image are capillaries and ascending and descending limbs. These medullary structures, which do not have significant active transport, were represented in the model by a source term, S_c , at each tissue point that takes into account the difference between oxygen delivery and consumption by these vessels and tubules. Without significant active transport in these structures in the inner medulla, it was assumed that they delivered more oxygen than they consumed, i.e., they provided a small net positive source of oxygen. Since they were not represented in the Pannabecker and Dantzler image, it was assumed that the capillaries and tubules were uniformly distributed within the tissue cross section, so that the source term, S_c , was assumed a fixed constant value at each tissue point. A value of $0.1 \mu\text{M/s}$ was used, based on simulations from a previous study [21]. We studied model sensitivity to this parameter in the Results (see Section 3.7).

2.3. Numerics

Because the model simulates oxygen transport in a two-dimensional cross section, axial changes in PO_2 along the vessels or collecting ducts were not represented, and luminal PO_2 was assumed known. Specifically, DVR, AVR, and collecting duct lumina were assigned fixed baseline PO_2 values of 40, 35, and 10 mmHg, respectively [8], corresponding to average values for these inner medullary tubules and vessels near the outer-inner medullary boundary (i.e., where the image was taken).

To compute the model solution, Eq. (1) was discretized using a centered finite differences method in space and solved with an explicit time-stepping scheme. The tissue cross section was discretized into a 49×49 grid, corresponding to a grid spacing of $4.25 \mu\text{m}$; a 1-ms time step was used. The solution was advanced to an approximate steady state, measured by a difference of less than 10^{-4} in L_2 -norm between solutions computed at successive time steps.

3. Results

Significant spatial variations in tissue PO_2 may arise from a number of factors. The location of the oxygen-carrying DVR outside of the collecting **duct cluster** may yield a higher PO_2 in the intercluster region. The high oxygen consumption of the collecting ducts may further lower the PO_2 within the clusters. In a set of simulations, we assessed the contribution of each of these factors to the generation of radial PO_2 gradients in the upper inner medulla.

3.1. Basal tissue O_2 consumption

To assess the impact of collecting duct active O_2 consumption, we first conducted simulations with only basal tissue oxygen consumption, i.e., active O_2 consumption was eliminated. Figure 3 shows the PO_2 relative frequency distribution in the four cases. The average PO_2 is comparable in the cases (28.8 and 29.4 mmHg for ES and EH, and 29.3 mmHg for AS and AH); however, the spread is larger in cases AS and AH (actual spacing), with a standard deviation of 6.2 mmHg compared to 4.8 and 5.0 mmHg in cases ES and EH (equal spacing). This leads to a lower minimum PO_2 and larger maximum PO_2 within the tissue region with actual-spaced vessels and collecting ducts. The differences among these cases are attributed to the variations in spacing (e.g., comparing AS and ES, or AH and EH).

3.2. With active tissue O_2 consumption

Next we incorporated collecting duct active O_2 consumption. Figure 4 shows the PO_2 frequency distribution with active O_2 consumption near the collecting duct wall, in addition to basal tissue O_2 consumption everywhere. Again, the average PO_2 is comparable in the four cases, with a larger spread in cases AS and AH. Inclusion of active O_2 consumption also leads to a lower average PO_2 everywhere.

Figure 5 is a contour plot of the tissue PO_2 in the cross section, in the four different cases. Heterogeneous spacing of inner medullary tubules and vessels creates more patches of tissue with low PO_2 (Figs. 5b and 5d) that are not present if the structure is assumed to be uniform (Figs. 5a and 5c). In particular, the clustering of the collecting ducts away from the DVR leads to areas with high oxygen consumption far from main oxygen supply. In addition, heterogeneous sizing of collecting ducts leads to a wider spread in PO_2 (more evident in Figs. 5a and 5c), though less than the effect of heterogeneous spacing.

3.3. Range of active O_2 consumption rates

Given the essential role of collecting duct active O_2 consumption in the generation of interstitial PO_2 variations, we conducted additional simulations to assess the sensitivity of

model predictions to changes in active O_2 consumption rate. This is particularly pertinent, given that collecting duct active NaCl transport rate is not well-characterized.

Figure 6a shows the effect of changing the active O_2 consumption rate on the minimum tissue PO_2 for the four cases. For the baseline maximum active O_2 consumption rate, 0.323 mM/s, the minimum tissue PO_2 is comparable in the four cases. As the maximum rate is increased, the separation between the cases widens. For moderate increases in consumption rate, the minimum tissue PO_2 is greater in the equal spacing cases than the actual spacing cases, regardless of collecting duct size, indicating that spatial arrangement has a stronger effect. For larger increases (rates higher than 1.9 mM/s), the minimum tissue PO_2 is greater in the equal collecting duct size cases than the heterogeneous collecting duct size cases, indicating that heterogeneity in collecting duct diameter has a stronger effect. If the maximum active O_2 consumption rate is ten times the base case, the minimum tissue PO_2 is essentially zero with actual spacing and collecting duct size, but over 3 mmHg with uniform spacing and collecting duct size.

Figure 6b shows how median tissue PO_2 changes with changing active O_2 consumption rate. For the baseline maximum O_2 consumption rate, the median tissue PO_2 is highest – at 30.7 mmHg – in the AS and AH cases, compared to 30.4 in the EH and 30.0 in the ES cases. This trend continues as the maximum rate increases, with all cases declining linearly.

The differences between the cases grow slightly with a maximum O_2 consumption rate ten times the baseline, with median PO_2 at 28.1 mmHg in the AS and AH cases, compared to 27.6 in the EH and 26.9 in the ES cases. Thus, the spread between the lowest and highest median PO_2 cases grows from 0.7 mmHg to 1.2 mmHg, an increase of 0.5 mmHg. This is compared to the spread between the lowest and highest minimum PO_2 case growing from 0 to 3.2 mmHg – six times larger.

3.4. Effects of lower DVR PO_2

Luminal PO_2 of the DVR and AVR were prescribed. To study the degree to which the predicted PO_2 values are sensitive to changes in vascular PO_2 , we conducted simulations in which we varied those PO_2 values. Figure 7 shows the effect of changing DVR (and AVR) PO_2 on median tissue PO_2 in the four cases. Like with changing maximum active tissue O_2 consumption rate, the AS and AH cases result in a larger median tissue PO_2 than the ES and EH cases, and this difference is maintained as DVR PO_2 declines. Here, the differences actually decline slightly, from 0.7 mmHg with DVR $PO_2 = 40$ mmHg versus 0.4 mmHg with DVR $PO_2 = 30$ mmHg.

Compared to changing the maximum O_2 consumption rate, changing DVR PO_2 causes the median tissue PO_2 to decline significantly faster. Changes in median tissue PO_2 between DVR $PO_2 = 40$ mmHg and DVR $PO_2 = 30$ mmHg were 8.0 (ES), 8.2 (EH), 8.3 (AS), and 8.3 (AH) mmHg in the four cases. This is compared to changes in median tissue PO_2 between maximum active O_2 consumption rates of 0.323 mM/s and 3.23 mM/s of 3.1 (ES), 2.8 (EH), 2.6 (AS), and 2.6 (AH) mmHg.

3.5. Effects of changing CD luminal PO_2

As in the DVR and AVR, CD luminal PO_2 was prescribed. Because this does not allow for changes in O_2 delivery from the collecting duct, which could have a significant effect on tissue oxygenation, we also assessed the extent to which tissue PO_2 was affected by variations in CD PO_2 . We found that due to the low maximum active O_2 consumption rate in the baseline case, minimum tissue PO_2 hovers around this prescribed CD PO_2 value (Results not shown).

As can be seen in Figure 6a, minimum tissue PO_2 begins to exhibit a strong dependence on maximum active O_2 consumption rate for larger rates. Thus, we further examined the effect on minimum tissue PO_2 of changes in prescribed CD PO_2 , for a maximum active O_2 consumption rate ten times baseline. Results of this simulation are shown in Figure 8.

Similarly to the results of Section 3.3, for a maximum active O_2 consumption rate ten times baseline, minimum tissue PO_2 is most strongly affected by heterogeneous collecting duct diameter, as the AH and EH cases have a lower minimum tissue PO_2 than the AS and ES cases, independent of CD PO_2 . Increasing prescribed CD PO_2 has the general effect of increasing minimum tissue PO_2 , as would be expected, but the general differences among the four cases is unaffected by CD PO_2 value, especially for values greater than 10 mmHg. However, in all four cases, for some CD PO_2 between 5 and 10 mmHg, the minimum tissue PO_2 falls to essentially zero, and stays there for further decreases in CD PO_2 .

3.6. Effects of non-uniform diffusivity

The non-uniform density of tubules and vessels in the inner medulla creates non-uniform barriers to diffusion in the tissue. In the baseline case, the diffusion coefficient, D , was assumed constant throughout the tissue region; however, areas of tissue with a higher density of obstacles to diffusion will have a lower effective diffusivity. Thus, we conducted simulations where D was varied within the tissue region according to the local density of tubules and vessels. Namely, D at a particular tissue point was scaled inversely to the number of tubules and vessels within a 10 μm radius of that point.

Figure 9 shows the PO_2 frequency distribution with the non-uniform diffusivity included, for the different cases. Compared to the base case, the frequency distributions are flattened in all cases, with decreased peaks. Further, the median tissue PO_2 in all cases is decreased, most significantly in the AS and AH cases, where median PO_2 drops by almost 20%, from 30.7 mmHg to 24.6 mmHg; this is compared to drops of just 3.7% and 4.9% in the ES and EH cases, respectively. Minimum tissue PO_2 also falls in all cases, to 8.1 mmHg in the AS and AH cases, and to 7.5 mmHg in the ES and EH cases.

3.7. Sensitivity to capillary source strength

The value of the capillary source strength, S_C , was fixed at a constant value of 0.1 $\mu\text{M/s}$ in the base case. Since basal O_2 consumption was assumed considerably larger than this value (and active O_2 consumption even larger than that), we wanted to examine the extent to which heterogeneity of tissue PO_2 was sensitive to increasing values of S_C . We thus conducted simulations in which S_C was increased in steps up to 500 $\mu\text{M/s}$.

Figure 10a shows the PO_2 frequency distribution as S_C is increased in the AH case. As S_C is increased from 0.1 to 10 $\mu\text{M/s}$, the distribution is essentially unchanged; thus, only $S_C = 10$ is plotted. As S_C is increased from 10 to 500 $\mu\text{M/s}$, the distribution expectedly shifts toward higher PO_2 values, as the oxygen delivery term is being increased. The spread, however, remains large throughout the tissue region, as the standard deviation actually increases from 6.48 to 7.22 to 9.80 to 13.0 mmHg, as S_C grows from 10 to 100 to 300 to 500 $\mu\text{M/s}$. Some of this spread can be attributed to the low fixed CD PO_2 at 10 mmHg in the model. Thus, we ran simulations where CD PO_2 was increased with increasing capillary source strength, by the amount over base case that increasing S_C caused the maximum tissue PO_2 to increase.

Figure 10b shows these results, again for $S_C = 10$ through $S_C = 500$ $\mu\text{M/s}$. As expected, this results in a larger shift of tissue PO_2 levels to higher values, and decreases the standard deviation of the distributions. The standard deviation for S_C values of 10, 100, 300, and 500 $\mu\text{M/s}$ in this case are 6.48, 6.10, 6.33, and 8.82 mmHg. Thus, there is actually a minor decrease in spread in going from an S_C value of 10 to 100 $\mu\text{M/s}$, indicating a parameter regime in which heterogeneity can slightly decrease with increasing capillary source strength. However, it should be noted that for values of $S_C \geq 100$ $\mu\text{M/s}$, the maximum tissue PO_2 is larger than the DVR PO_2 , which is not a physiological state. In general, for all values of S_C , the trend is that the standard deviation is maintained in all cases, indicating the robustness of the heterogeneity in tissue PO_2 over a significant range of capillary oxygen delivery.

4. Discussion

We have developed a detailed model of oxygen transport in a cross section of the upper inner medulla of the rat kidney. The positions and dimensions of tubules and vessels were based on an immunohistochemical localization study by Pannabecker and Dantzler [12]. Model simulations suggest that owing to the metabolic demands of the active NaCl reabsorptive process of collecting duct epithelia, and to the separation of the collecting ducts from the oxygen-carrying descending vasa recta, oxygen tension in the interstitial fluid surrounding the collecting ducts is substantially lower than that around the descending vasa recta (10–18 vs. 33–39 mmHg).

In the rat renal inner medulla, PO_2 was estimated at 25 mmHg by Liss et al. [22], at a depth of 1–2 mm below the outer-inner medullary boundary. In human kidney, Leonhardt and Landes [23] measured a PO_2 of 26 mmHg at the outer-inner medullary boundary and 22 mmHg at 3 mm below the boundary. One study of dog kidney measured a PO_2 of ~ 4 mmHg in the inner medulla [24], and another of 8–10 mmHg at 0 to 2 mm below the outer-inner medullary boundary [25]. Simulation results from the present study generally lie within this range, with median PO_2 values between 22 and 31 mmHg. PO_2 values near those measured in the dog kidney are achieved for larger maximum active O_2 consumption rates.

Previous modeling studies have attempted to address the impact of heterogeneous vessel and tubule structure on functional properties in tissues ranging from skeletal muscle [26, 27, 28, 29] to kidney [8, 9, 30, 10, 31, 32, 33, 34, 35]. In skeletal muscle, Lo et al. [28] used explicit image data from cross sections to analyze oxygen transport in a 2D tissue slice. A similar

approach to this skeletal muscle analysis was used in the present study to examine oxygen transport in the inner medulla of the kidney, including a finite-difference method for spatial discretization. 3D models of skeletal muscle oxygen transport from Goldman et al. [27] and Tsoukias et al. [29] used morphological parameters to construct distributions of vessels within a tissue block, but did not use explicit image data to get at heterogeneity of vascular structure. More recently, models of metabolic blood flow regulation in the skeletal muscle microcirculation from Roy et al. [36] and Fry et al. [26] used high-resolution images of microvascular networks to explicitly represent positions of vessels within a volume of tissue. These studies involving structural heterogeneity have been strongly guided by available images in certain types of more easily-accessible muscle tissue. More explicit representations of kidney tubular and vascular geometry have been hindered by a relative lack of high-resolution imagery until recently. Thus, many models of oxygen transport in the kidney include an idealized or simplistic representation of the spatial arrangements of tubules and/or vessels. For example, Gardiner et al. [31] developed a model of oxygen shunting between vessels in the renal cortex. That model does not represent the actual radial positions (perpendicular to flow directions) of the vessels, but instead simulates oxygen transport between pairs of arterial and venous vessels with different lumen separation distances and degrees of wrapping, one pair at a time.

Chen et al. [8, 9] developed a highly detailed oxygen transport in the outer medulla of the rat kidney. The spatial arrangement of tubules and vessels is represented using a “region-based approach.” Four interconnected regions are included in the model, each representing the interstitial fluid and interstitial cells of a section of the outer medulla. At each medullary level, the fluid composition in each region is assumed to be homogeneous. Each tubule or vessel is assigned to one of the (four) regions and interact only with fluid in that region. That formulation captures some of the preferential interactions between tubules and vessels that arise from their relative positions, but because only four regions are represented, the representation of the radial distribution of the tubules and vessels is coarse. Also, the physical dimensions of each class of tubules (e.g., collecting ducts) are assumed to be uniform.

In contrast, the present study incorporates a faithful 2D representation, based on an image from Ref. [12], of a section of the cross section of the rat kidney. Multiple collecting ducts are represented, each with different physical dimensions and position. Thus, the present study yields much richer information about the impacts of the radial tubular and vascular arrangements on oxygen transport. Indeed, simulation results reveal significant localized variations in PO_2 , which arise from heterogeneous spacing and dimension of tubules. Those variations would not have been revealed in a simpler model with idealized geometry. In a recent editorial [37], Weinstein predicted that the structure of the inner medulla would have only a small effect on Na^+ and urea radial concentration gradients, due to short diffusion distances, but that O_2 could sustain such a gradient because of O_2 consumption in medullary tissue. Results of the present study support the latter half of this hypothesis.

Nonetheless, in some aspects the present model is more limited compared to the study by Chen et al. [8, 9]. Most notably, the present model represents a cross section of the renal medulla, whereas Chen et al. model the entire outer medulla. Consequently, axial flows are

not represented in the present model, and PO_2 in the lumens of the collecting duct and vasa recta are assumed known *a priori*. Those luminal PO_2 values are estimated based on experimental data and model simulations [25, 21, 23, 22]. But, as our simulations show, medullary oxygenation is sensitive to variations in vascular PO_2 . In addition, as seen in Figure 8, a limitation of the model is that due to the prescription of a constant PO_2 , the collecting ducts provide a source of oxygen even at high rates of O_2 utilization, where minimum tissue PO_2 falls to zero. Thus, it is a worthwhile exercise to extend the present model to a full 3D model of medullary oxygenation, although the computational cost of such a model will likely be high.

Inner medullary collecting ducts are believed to mediate substantial active NaCl reabsorption, based on functional and modeling studies [19, 32, 38, 15]. However, the rate of active NaCl transport, or the rate of active O_2 consumption, in the inner medullary collecting duct is not well-characterized. Estimates from these experiments and modeling studies give a range of at least an order of magnitude. In a study of inner medullary collecting duct transport by Weinstein [15], sodium flux values from a range of investigators were collected and summarized. The fluxes ranged from 0.25 to 17.5 nmol/cm²/s, over a variety of experimental conditions. Using our baseline maximum active O_2 consumption rate of 0.323 mM/s, and assuming that 12 moles of Na^+ are reabsorbed though the transcellular pathway per mole of O_2 consumed [21], we estimate a maximum active sodium transport rate of 5.3 nmol/cm²/s. This is consistent with the experimental estimates; this baseline is actually at the lower end of the estimates, as our estimated rate is a maximum, when concentrations are not limiting, which would not be the case *in vivo*. When a range of increasing values for maximum active O_2 consumption rate was examined in the present study, simulation results indicated that the effects of structural heterogeneity in the inner medulla are exacerbated as oxygen consumption increases (Fig. 6a). For smaller values of maximum active consumption rate, minimum tissue PO_2 levels were maintained around 10 mmHg in all cases. However, for values in the upper range, the differences between the cases were drastic. Both heterogeneity in spatial location of tubules and vessels as well as CD size contributed to a noticeable decrease in the minimum PO_2 , even falling below 1 mmHg in the actual spacing and CD size case. This indicates that patches of tissue may be exposed to hypoxia, unless tubular function is maintained via glycolysis.

Importantly, this increase in hypoxic regions may not be reflected in measurements of average or median tissue PO_2 . Simulations indicate that large changes in minimum tissue PO_2 are not necessarily accompanied by correspondingly large changes in median tissue PO_2 (Figure 6). In fact, the case with the largest median tissue PO_2 (AH) tended to have the smallest minimum tissue PO_2 for most consumption rates. These results strongly suggest that much care must be taken in drawing conclusions from average tissue PO_2 measurements when it comes to the health of renal tissue, due to the structural heterogeneity in the spatial arrangement of the inner medulla.

Model simulation results also indicate that median PO_2 is much more sensitive to DVR PO_2 (O_2 delivery) than to maximum O_2 consumption rate (O_2 demand). Oxygen delivery to the inner medulla can vary under a number of physiological and pathophysiological conditions. For example, elevated medullary blood flow, taken in isolation, would raise DVR PO_2 in the

inner medulla, and vice versa. Medullary blood flow is known to be modulated by vasopressin [39, 40, 41, 42] and to increase when renal interstitial hydrostatic pressure is elevated [43]. Another example is renal hemodynamics during cardiopulmonary bypass surgeries: the anesthetics lower blood pressure and medullary blood flow, assuming that the latter is not perfectly regulated. Furthermore, systemic hematocrit is drastically reduced, although the impact on oxygen supply is partially compensated by highly oxygenated plasma. Together, lower mean arterial pressure and hemodilution likely results in a fraction of cardiopulmonary bypass patients suffering from acute kidney injury post-surgery.

In summary, model results suggest that the heterogeneous spatial distribution of the tubules and vessels in the upper inner medulla of the rat kidney may give rise to substantial spatial variations in interstitial PO_2 . This finding may help explain the occurrence of certain cases of renal hypoxic injuries.

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Figure 1.

Image of inner medullary cross section reprinted from Pannabecker and Dantzler [12], on which the model structure is based (white space in top left corner was used to label the image in [12]). Included are spatial locations of DVR (green), AVR (red), and CD (blue). Scale bar, 30 μm .

(a) Equally-spaced tubules/vessels. (b) Actual-spaced tubules/vessels.

Figure 2.
Spatial locations of DVR (green), AVR (red), and CD (blue).

Figure 3.

Relative frequency distribution of PO_2 in the different cases, with only basal tissue O_2 consumption. (Plots of AS and AH cases were indistinguishable in the figure, so both are represented by the “Actual spacing” curve.)

Figure 4.

Relative frequency distribution of PO_2 in the different cases, with basal and active tissue O_2 consumption. (Plots of AS and AH cases were indistinguishable in the figure, so both are represented by the “Actual spacing” curve.)

(a) Equal spacing, Same-sized CDs (ES) (b) Actual spacing, Same-sized CDs (AS)

(c) Equal spacing, Hetero CD sizes (EH) (d) Actual spacing, Hetero CD sizes (AH)

Figure 5.

PO_2 distribution in inner medullary cross section in all four cases, from 40 mmHg (red) down to 10 mmHg (blue). Includes basal O_2 consumption in all tissue and active consumption near CD wall.

(a)

(b)

Figure 6.

Minimum (a) and median (b) tissue PO_2 (mmHg) versus maximum active O_2 consumption rate (mM/s) in the four cases.

Figure 7.
Median tissue PO_2 (mmHg) versus DVR PO_2 (mmHg) in the four cases.

Figure 8.

Minimum tissue PO_2 (mmHg) versus CD PO_2 (mmHg) in the four cases, with maximum active O_2 consumption rate ten times baseline.

Figure 9.

Relative frequency distribution of PO_2 in the different cases, with non-uniform diffusivity. (Differences between AS and AH cases, as well as ES and EH cases, were nearly indistinguishable in the figure, so AS and AH are represented by the “Actual spacing” curve, and ES and EH are represented by the “Equal spacing” curve.)

(a)

(b)

Figure 10.

Relative frequency distribution of PO_2 in the AH case, for values of capillary source strength S_C of 10, 100, 300, and 500 $\mu\text{M/s}$. (a) Without and (b) with CD PO_2 adjusted to S_C increases.