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Assessment of Cadaveric Organ Viability During Pulsatile Perfusion Using Infrared Imaging

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Abstract

Assessment of pulsatile perfusion (PP) is limited to measurements of flow (V) and resistance (R). We investigated infrared (IR) imaging during PP as a means for precise organ assessment. IR was used to monitor 10 porcine kidneys during 18 hr of PP in an uncontrolled Donation after Cardiac Death model. An IR camera (Lockheed Martin) was focused on the anterior surfaces of the kidneys. The degree of temperature homogeneity was compared with standard measurements of V and R. IR thermal images correlated with V and R ($R=0.92, P<0.001$). IR detected an increase in homogeneity during PP by comparing standard deviation differences before and after PP ($P=0.002$), which was not evident by standard measurements of V and R. Finally, IR assessment allowed for measurement of dynamic changes in perfusion.

Keywords

Infrared imaging; Pulsatile perfusion; Viability

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Expanded criteria donors and donation after cardiac death donors have become a larger percentage of the donor pool, as transplant physicians try to meet the increasing demand for organ allografts. Many clinical centers have chosen to use machine pulsatile perfusion (PP) of kidneys from nonideal donors to evaluate and resuscitate these organs in efforts to maximize organ use and recipient outcomes (1–5). While studies support the utility of PP, current methods to evaluate the physiologic response to PP in vitro are essentially limited to measures of flow and resistance. However, changes in gross perfusion parameters during preservation may not accurately assess the overall functional state of the organ and, therefore, are not always determinative of response to PP or predictive of posttransplant renal function. Infrared (IR) imaging is based on the two-dimensional mapping of temperature differences by detecting natural emissions from tissue that are warmer (or cooler) than surrounding structures. With this imaging, assessments of tissue perfusion can be determined and measured. (6) We have previously used IR imaging to assess critical renal ischemia (7) and have now expanded these studies to address renal status during PP.

In undertaking this study, our hypothesis was that IR imaging of temperature could provide, at a minimum, the equivalent amount of prognostic data as current resistance and flow measurements, and provide additional dynamic assessments of perfusion. In this study, we demonstrate that IR imaging assessments of renal temperature have acceptable correlation with flow and resistance readings and can provide objective data regarding the homogeneity of flow within the kidney cortex. In addition, point differences in the flow dynamics throughout the kidney can be demonstrated by a moderate increase in perfusion pressure. The eventual ability to perform these measurements in real-time may allow for more accurate assessment of renal status during PP, allow for interventions to reduce reperfusion injury, and ultimately provide important prognosticators of subsequent graft function after transplantation.

METHODS

Kidney Recovery and Perfusion

Under IACUC approval (NIHUOB006) 10 pig kidneys (5 pairs) were recovered from a regional USDA slaughterhouse. After 30 min of warm ischemia, kidneys were initially flushed with a liter of 4°C Lactated Ringers solution followed by 4°C Modified Belzer Machine Preservation Solution (Transmed Corporation, Elk River, MN). The perfusion cassette was primed with Modified Belzer Machine Preservation Solution and cooled to 5°C. The cassette with two kidneys was installed and connected onto a Waters system (RM3, Waters Medical Systems, Rochester, MN). The perfusion system's pulse rate was adjusted to 1 pulse/sec, and perfusate temperature (°C), pressure (mm Hg), flow (mL/min), and resistance (mm Hg/mL per minute) were recorded and analyzed off-line.

IR Imaging

An advanced digital IR camera (Infrared Focal Plane Array Camera, Lockheed Martin IR Imaging Systems, Goleta, CA) was used to image both perfused kidneys' local temperature gradients simultaneously by passively detecting the IR emissions. The camera has a sensitivity of 0.02°C and images (320×256 pixels each) were obtained at 5 sec intervals and digitized at 14 bits. The IR camera was installed onto a modified surgical microscope stand approximately 100 cm above the cannulated kidneys. The IR camera was focused on the renal parenchyma during the time of the perfusion experiment. IR images were acquired during several hours of kidney resuscitation, visualized in real time and stored for further off-line image analysis.

Offline Data Analysis

Temperature profiles were extracted from successive image sets (ENVI/IDL software, ITT Visual Information Solutions, Boulder, CO) and compared with resistance and flow profiles. The mean adjusted IR temperature readings for whole kidney and regions of interest (ROIs) were compared with the renal resistance and renal flow. Regression analysis was performed (Systat Software, Inc., Richmond, CA) to reveal the relationships between adjusted IR temperature and renal resistance, as well as between adjusted IR temperature and renal flow. For each original temperature profile, the derivative value was calculated using the following formula: $y'(t) = [y(t + h/2) - y(t - h/2)]/h$. The degree of homogeneity of kidney perfusion was assessed using standard deviation of IR temperatures for the entire kidney ROI. Paired Student's *t* test was applied to compare the mean temperature and standard deviation of temperature for the entire kidney.

RESULTS

IR Correlates With Standard Measures of Flow and Resistance

The IR-derived temperature values measured were found to have a strong direct correlation with measured resistance within the kidney (Fig. 1A, $R = 0.92$, $P < 0.001$). In addition, there was an indirect correlation with flow (Fig. 1B, $R = 0.92$, $P < 0.001$). Finally, the derivative of the temperature (as calculated by slope assessment) also strongly correlated with renal resistance ($R = 0.77$, $P < 0.001$). These data suggest that the IR measurement of temperature can give the same amount of prognostic data as current resistance and flow measurements.

IR Detects an Increase in Homogeneity During PP

After confirming the correlations between flow and resistance to IR-measured temperature, our attention turned to the manner in which IR temperature measurements can increase the amount and utility of data obtained during PP. We evaluated the amount of homogeneity of temperature within the perfused renal allograft. During PP, the mean temperature of the kidneys decreased significantly ($n = 6$, $P = 0.002$). In addition, the standard deviation of temperature also decreased during the perfusion period ($n = 6$, $P = 0.03$), suggesting an improvement in homogeneous flow within the organ (Fig. 2).

IR Can Measure Dynamic Changes in Perfusion

Finally, we attempted to determine the level of feedback that IR assessment provides by sharply increasing the perfusion pressure. We selected a 60-min perfusion run during which an attempt was made to interrogate the local response to a bolus infusion. After 20 min of steady state perfusion with 27 mm Hg, we changed the perfusion pressure abruptly, with a pressure increase of 24 mm Hg for the next 22 min, followed by a decrease of 16 mm Hg, and measured the thermal response over the subsequent hour.

Figure 3 shows synthesized images, with each pixel representing the calculated temperature derivative. Temperature derivative images were synthesized for selected representative time points: steady state at 17 min (Fig. 3A), the increases in pressure at 21 min (Fig. 3B) and 39 min (Fig. 3C), and after the decrease in pressure at 43 min (Fig. 3D). The entire perfusion period was not assessed due to the current limitations of image acquisition and analysis. The IR temperature derivative image was homogeneous (Fig. 3A) before the increase in mean pressure at 17 min. The IR temperature derivative image became heterogeneous and showed large negative derivative values (Fig. 3B,C) and positive derivative values (Fig. 3D) after the pressure changes. This enables IR to be used to assess dynamic responses during perfusion throughout an entire organ.

DISCUSSION

The results of this series of studies in the evaluation of kidneys undergoing PP demonstrate that IR is able to provide data that correlate with the standard measures of flow and resistance. Figure 1 demonstrates the excellent correlation of resistance and flow with temperature and the derivative of temperature. In addition, IR can provide data on the homogeneity of flow within the renal allograft, which standard measures of flow and resistance are unable to provide. The homogeneity of flow can be quantified by measuring the standard deviation of temperature measured throughout the kidney as shown in Figure 2 or by generating a visual temperature derivative map of the renal allograft, as shown in Figure 3.

The use of IR during PP offers several potential advantages over standard measures. Detection of inhomogeneous flow can allow for the use of pharmacologic agents during PP to increase homogeneity that could lead to less vasospasm within the graft after reperfusion and potentially decrease delayed graft function. As such renal allografts that do not respond to pharmacologic manipulation may be allografts that should not be implanted due to a higher likelihood of primary nonfunction. In addition, by perturbing the system at steady state a dynamic measure of perfusion response within the renal allograft can be measured in real time and viewed as a temperature derivative map (Fig. 3A–D). This study is limited by the fact that subsequent renal function was not evaluated and the current methodology to assess perfusion on a pixel-by-pixel basis as, in Figure 3, requires off-line analysis. We are addressing these two issues with a planned transplant study and increased computing power for real-time assessment of homogeneity.

IR imaging of porcine kidneys under PP demonstrates that this technique provides the same level of data as current prognosticators of organ function on PP. The data also demonstrate that IR imaging provides valuable spatial information on kidney perfusion heterogeneity. Such imaging can be used to refine our ability to predict PP outcomes and to use the images as real-time guidance for different intervention procedures on the prospective graft. The effect of inhomogeneous flow on graft function needs to be substantiated in both PP cadaveric kidneys and living-related donors using several techniques (open, hand-assisted, and laparoscopic).

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FIGURE 1.

Linear relationships between adjusted IR temperature and renal resistance (A) and flow (B). Assuming default perfusate temperature as 5°C and default temperature of cassette external wall as 15°C, pixel intensity of IR images were adjusted to eliminate the effects of the environment temperature fluctuations via the equation: adjusted temperature = real temperature + (5°C–real perfusate temperature) + (15°C–real temperature of cassette external wall).

FIGURE 2.

Mean temperature (A) and its standard deviation (B) for a whole kidney before and after PP (n=6, pair Student's *t* test, where * represents a significant difference, $P<0.05$).

FIGURE 3.

Temperature derivative images synthesized for steady state condition at 17 min (A), at 21 min (B), and 39 min (C) during mean pressure elevation, and at 43 min during pressure decrease (D). Note: to show heterogeneity effectively, the ranges of derivative values applied to images “A” and “B” are different than those applied to “C” and “D.”