

## Effect of Isoflurane on Aortic Impedance in Mice

Anilkumar K. Reddy<sup>1</sup>, *Senior Member IEEE*, Khodidad Namiranian<sup>2</sup>, Eric E. Lloyd<sup>2</sup>,  
Robert M. Bryan, Jr.<sup>2</sup>, George E. Taffet<sup>1</sup>, and Craig J. Hartley<sup>1</sup>, *Senior Member, IEEE*

**Abstract**—Isoflurane is the most commonly used anesthetic in mice. We studied the effect of low and high levels of isoflurane (also a potent coronary vasodilator) on aortic impedance in mice. Aortic impedance was determined using pressure and flow velocity signals at baseline (B, pentobarbital anesthesia), low (Iso1, 1%), and high (Iso2.5, 2.5%) levels of isoflurane. Significant differences were observed in peak and mean flow velocities, systolic, diastolic, mean and pulse pressures at B and Iso2.5. However in impedance indices only peripheral vascular resistance was significantly different. No changes were observed in the harmonic components that represent pulsatile characteristics of the aorta. Peak left ventricular (LV) pressure was significantly lower at Iso2.5 when compared to B, but  $\pm dP/dt$  and  $\tau$  (time constant of LV relaxation) did not change significantly indicating that LV contractility was unaffected. These results show that various levels of isoflurane cause significant changes in vascular hemodynamics and care must be taken to minimize these differences when using isoflurane as an anesthesia.

**Keywords**—Aortic impedance, Aortic pressure, Aortic flow velocity, Anesthesia, LV contractility.

### I. INTRODUCTION

EXPERIMENTAL studies to measure cardiovascular signals in mice require anesthetizing these animals in many cases. One of most commonly used anesthetic is isoflurane gas, usually administered in combination with either oxygen or room air. It is well known that any anesthesia affects mouse hemodynamics and isoflurane is no exception. Previously we showed that isoflurane is potent coronary vasodilator [1]. Other investigators reported that there is a general decrease in the heart rate and other cardiac indices when anesthesia is administered to conscious mice [2] but the influence of various levels of isoflurane on arterial hemodynamics in mice anesthetized with pentobarbital sodium is not known. Here, we studied the effect of low and high levels of isoflurane on aortic impedance in mice. Aortic impedance is typically calculated using pressure and flow signals. However to determine aortic impedance independent of body weight or size, we used velocity instead of flow [3].

Manuscript received April 23, 2009. This work was supported in part by NIH grants R01-HL22512, P01-HL42550, R01-AG17899, R41-HL76928, and K25-HL73041. Authors Namiranian, Lloyd, and Bryan, Jr., are with the Department of Anesthesiology at Baylor College of Medicine. Authors Reddy, Taffet, and Hartley are with the Department of Medicine, Section of Cardiovascular Sciences, Baylor College of Medicine and The Methodist DeBakey Heart & Vascular Center, Houston, TX 77030. areddy@bcm.edu.

### II. METHODS

Five wild type mice (aged 2 months) were used in the study. Mice were anesthetized with pentobarbital sodium given intraperitoneally (4 mg/ml, 10  $\mu$ l/g body weight of mouse). The neck and xiphoid areas were shaved and the mouse was placed in a supine position with its paws taped to electrodes on an ECG board. The right carotid artery of the mouse was isolated, tied off distally, and the proximal end was temporarily closed. A small cut was made in the artery and a 1F Millar catheter (SPR1000, Millar Instruments, Inc., Houston, TX) was inserted and was held in place with a suture tied over the artery-catheter overlap region. The proximal end of the artery was then opened and the catheter was advanced into the aorta. Aortic blood velocity was measured with a 20 MHz pulsed Doppler probe placed externally with its sample volume ranged to be at the same location as the pressure sensor in the aorta [3]. We measured aortic flow velocity and blood pressure in each anesthetized mouse (baseline B) and with interventions of 1% isoflurane (Iso1) mixed with oxygen and 2.5% isoflurane (Iso2.5) mixed with oxygen as interventions. Then the catheter was advanced into the LV. Again, pressure was measured at B, Iso1 and Iso2.5. A two-second long segment of blood pressure and quadrature Doppler audio signals was acquired by a Doppler signal processing workstation (Indus Instruments, Houston, Texas). Peak velocity envelope and blood pressure signals were extracted from the stored data file, and a DFT was performed on each signal to obtain magnitude and phase spectra. The modulus of input impedance was then calculated as  $|Z_i| = |P|/|V_{avg}|$ , where  $|P|$  and  $|V_{avg}|$  are the pressure and average luminal velocity moduli, respectively. From impedance modulus, peripheral vascular resistance ( $Z_0$ ), impedance at 1<sup>st</sup> harmonic ( $Z_1$ ), and characteristic impedance ( $Z_C$ , average of 2<sup>nd</sup>-10<sup>th</sup> harmonics) were calculated [4]. Systolic (ABP<sub>sys</sub>), diastolic (ABP<sub>dias</sub>), mean (ABP<sub>mean</sub>) and pulse (APP) pressures were obtained from aortic pressure signals. Peak (PeakAFV) and mean (MeanAFV) velocities were obtained from aortic velocity signals. Peak pressure (PeakLVP),  $\pm dP/dt$ , and  $\tau$  (tau) were calculated from LV pressure signal. Data are presented as mean $\pm$ SEM.

### III. RESULTS

The results are summarized in Table I. We found that the heart rate did not change significantly from B at Iso1 or Iso2.5. In general pressures decreased and flow velocities increased with interventions of isoflurane with greater changes occurring at Iso2.5 when compared to B. PeakAFV

and MeanAFV were higher and ABPsys was lower at Iso1 when compared to B. While  $Z_1$  and  $Z_c$  were unchanged  $Z_0$  decreased significantly from B to Iso1, and from Iso1 to Iso2.5. Pulse wave velocity (PWV) was calculated using the equation  $Z_c = \rho \cdot PWV$ , where  $\rho$  is the density of blood [3].

TABLE I  
Summary of hemodynamic data from 5 WT mice.

Parameter	Baseline	Iso1	Iso2.5
$Z_0$ (dynes-s/cm <sup>3</sup> )	9386±489	7733±591†	6315±433‡*
$Z_1$ (dynes-s/cm <sup>3</sup> )	654±45	640±36	654±51
$Z_c$ (dynes-s/cm <sup>3</sup> )	306±30	284±20	333±27
PWV= $Z_c/\rho$ (cm/s)	281±28	261±18	306±25
PeakAFV (cm/s)	111.1±5.8	124.2±7.0	136.1±6.6‡*
MeanAFV (cm/s)	23.5±0.9	27.2±1.6	29.4±1.0‡
ABPsys (mmHg)	97.1±5.4	93.1±4.9	86.2±2.4‡
ABPdias (mmHg)	70.1±3.5	63.2±3.8†	53.3±2.2‡
ABPmean (mmHg)	83.4±4.3	78.0±4.3†	69.1±2.0‡
APP (mmHg)	27.0±1.9	29.9±1.7†	32.9±2.3‡*
HR (beats/min)	417±21	423±20	414±28
PkLVP (mmHg)	102.2±2.9	99.2±3.6	94.8±3.0‡*
LVEDP (mmHg)	7.1±0.9	7.2±0.4	6.9±0.8
+dP/dt (mmHg/s)	9798±1481	8154±599	7678±218
-dP/dt (mmHg/s)	-13445±3408	-13209±2905	-10644±1673
tau (msec)	7.4±0.9	7.5±1.3	8.9±0.9

Values are mean±SEM; PWV - pulse wave velocity;  $\rho$  - blood density  $\approx 1.06$  g/cm<sup>3</sup>

† -  $p < 0.05$  Iso1 vs Baseline; ‡ - Iso2.5 vs. Baseline; \* - Iso1 vs. Iso2.5

#### IV. DISCUSSION

The gas, isoflurane is now widely used to anesthetize mice during experimental studies but it is not as widely recognized as a potent coronary vasodilator in mice. We previously reported that coronary flow velocity in mice increases as much as 3 times when the administered level of isoflurane (mixed with oxygen) is increased from 1% to 2.5% [1,4]. However, how the various levels of isoflurane affect arterial hemodynamics. In order to delineate the effects of isoflurane levels we used pentobarbital sodium as a baseline anesthesia and used Iso1 and Iso2.5 as interventions. In doing so, we made the assumption that the interaction between pentobarbital sodium and isoflurane is minimal.

The results showed that impedance values were comparable to those previously reported by us [3]. Also, the values of PWV calculated using  $Z_c$  are comparable to those reported by us in mice [3,5]. While significant changes occur in aortic velocity and pressure with the administration of 2.5% isoflurane, aortic impedance indicates that the pulsatile component of the aorta is unchanged. However, the resistance of the peripheral vascular network seemed to decrease significantly. This means that the peripheral circulation is enhanced. Left ventricular pressure decreased significantly at Iso2.5 but there were no significant changes in contractility (+dP/dt) or relaxation (-dP/dt) of LV.

#### V. CONCLUSION

This results of this study shows that higher levels of isoflurane cause significant decreases in the resistance of the peripheral vascular system. So, one has to carefully consider the choice of other anesthetic agents instead of isoflurane when conducting perfusion studies in myocardium or other tissues.

#### ACKNOWLEDGMENT

The authors wish to acknowledge the contributions of Thuy Pham, Jennifer Pocius, Ross Hartley, and James Brooks for technical, surgical, and editorial assistance.

#### REFERENCES

- [1] C. J. Hartley, A. K. Reddy, S. Madala, L. H. Michael, M. L. Entman, and G. E. Taffet, "Effects of isoflurane on coronary blood flow velocity in young, old, and ApoE<sup>-/-</sup> mice measured by Doppler ultrasound," *Ultrasound Med. Biol.*, vol. 33, pp. 512–521, 2007.
- [2] B. J. Janssen, T. De Celle, J. J. Debets, A. E. Brouns, M. F. Callahan, and T. L. Smith, "Effects of anesthetics on systemic hemodynamics in mice," *Am. J. Physiol. Heart Circ. Physiol.*, vol. 287, pp. H1618–H1624, 2004.
- [3] A. K. Reddy, Y-H. Li, T. T. Pham, L. N. Ochoa, M. T. Treviño, C. J. Hartley, L. H. Michael, M. L. Entman, and G. E. Taffet, "Measurement of aortic input impedance in mice: effect of age on aortic stiffness," *Am. J. Physiol. Heart Circ. Physiol.*, vol. 285, pp. H1464–H1470, 2003.
- [4] C. J. Hartley, A. K. Reddy, S. Madala, L. H. Michael, M. L. Entman, and G. E. Taffet, "Doppler estimation of reduced coronary flow reserve in mice with pressure overload cardiac hypertrophy," *Ultrasound Med. Biol.*, vol. 34, pp. 892–901, 2008.
- [5] C. J. Hartley, G. E. Taffet, L. H. Michael, T. T. Pham, and M. L. Entman, "Noninvasive determination of pulse-wave velocity in mice," *Am. J. Physiol. Heart Circ. Physiol.*, vol. 273, pp. H494–H500, 1997.