An Initial Application of Transesophageal Doppler Echocardiography in Experimental Small Animal Models

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This study examined whether an intracardiac echocardiography catheter could be used for transesophageal echocardiography (TEE) examinations in normal rats, and intraoperative TEE in small animal models of disease. The study used 30 Sprague-Dawley normal rats, 10 rats undergoing coronary artery ligation, and 10 rats with experimentally induced mitral regurgitation. The rats were anesthetized with isoflurane and intubated. An intracardiac echocardiographic catheter was inserted into the esophagus. M-mode, 2-dimensional, and Doppler studies were performed in multiple views. TEE probe insertions were successful in all animals. Intraoperative TEE was safely performed in the rat models of myocardial infarction or mitral regurgitation. Mitral regurgitation was well assessed using color Doppler and pulmonary venous flow. This study demonstrates that TEE (including intraoperative TEE) can be safely performed in rats using an intracardiac echocardiographic catheter. It provides a new approach to the assessment of cardiac function and valvular regurgitation in small animals. (J Am Soc Echocardiogr 2005;18:626-31.)

Recent advances in genomics, and molecular and cell biology, have led to the widespread use of small animal models of human disease. One of the limitations in the use of small models is the need to kill large numbers of animals for ex vivo tissue and molecular analysis. Development of small animal imaging systems or adaptation of existing clinical devices might allow serial noninvasive investigation of biologic processes in vivo. Transesophageal echocardiography (TEE) provides high quality images of the heart and great vessels without the acoustic barriers of the ribs, skeletal muscles, or lungs. It has proved to be very useful for assessing cardiac structure and function, and valvular disease, in the clinical setting.[1-5] TEE can also be carried out intraoperatively.[6,7] Previous studies successfully used an intravascular ultrasound (IVUS) catheter (3.5F, 1.2-mm diameter) to image the rat heart through the esophagus.[8-12] These studies showed that 2-dimensional (2D) imaging of the heart could be obtained using an IVUS catheter in small animals.

Recently, a new phased-array intracardiac echocardiographic (ICE) catheter (AcuNav, Acuson-Siemens Corp, Mountain View, Calif) was developed. It has been used for ICE imaging in human beings and large animals.[13-16] Compared with the conventional IVUS catheter, the ICE catheter has spectral Doppler and color Doppler functions, allowing the assessment of hemodynamic and valvular function, which could not be obtained in the previous studies using the IVUS catheter. However, the ICE catheter (10F, 3.3-mm diameter) is larger than the IVUS catheter. The safety of the ICE catheter for TEE studies in small animals has not been completely established. A prior study evaluated the feasibility of the ICE catheter for TEE examinations in rabbits.[17] It showed that TEE studies could be safely performed in rabbits weighing more than 600 g. It remains unknown whether a satisfactory TEE study would be performed in smaller animals such as a rat weighting less than 600 g. Furthermore, the intraoperative application of the ICE catheter in small animal models required validation.

METHODS

Animal Preparation

The study protocol was approved by the institution's animal care and use committee. This institution has an animal welfare assurance on file with the Office for

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Protection from Research Risks, the National Institutes of Health (assurance No. A3045-01). The study was performed according to the guidelines of the American Physiological Society.

In all, 50 Sprague-Dawley rats (180-660 g, mean 307 ± 74 g) were used. Among them, 30 were normal. A total of 10 rats underwent surgical coronary artery ligation and 10 rats were given mitral regurgitation (MR). The rats were anesthetized with isoflurane (induction, 5% isoflurane and 95% molecular oxygen; maintenance, 2% isoflurane and 98% molecular oxygen) and intubated. The rats were placed on a ventilator (Hallowell AWS, Pittsfield, Mass) to maintain ventilations. Heart rate and oxygen saturation were monitored using respiratory gas monitor (Ohmeda 5250 RGM, Helsinki, Finland) throughout the study.

Animal Models

In all, 10 rats underwent coronary artery ligation to create a rat model of heart failure caused by myocardial infarction. Another 10 rats underwent a mitral valve procedure to develop a rat model of MR. An ICE catheter was inserted into the esophagus before the rat's chest was opened. Intraoperative TEE examinations were performed before, during, and immediately after operation. After the operation the ICE catheter was withdrawn and rats were returned to our animal facility for recovery.

Equipment

The study used a 10F ICE catheter equipped with a transducer at the tip of the catheter. This transducer has a 64-element phased-array oriented in a longitudinal plane with multiple frequencies from 5.5 to 10 MHz with maximal tissue penetration of 12 cm. The transducer generates M-mode, 2D, pulsed wave Doppler, color Doppler, and Doppler tissue images. The catheter tip is easily manipulated anteriorly, posteriorly, and laterally using the control knobs at the end of the catheter. The catheter is connected to a commercial echocardiograph (Sequoia 256, Acuson-Siemens Corp). All the study images were stored digitally and on VHS tapes. The ICE catheter was marked at 12-cm distance from the tip in our laboratory to measure the depth of the catheter insertion.

Probe insertion. Rats were placed in a supine position after intubation. After lubrication, the catheter was carefully inserted into the esophagus dorsal to the tracheal intubation tube. The catheter was slowly advanced. If significant resistance was encountered, the rat and intubation tube were slightly repositioned. Real-time 2D images were obtained to guide the position and the depth of the catheter. Probe insertion usually took less than 2 minutes.

Image Acquisition

2D. A series of 2D images were obtained with the probe in the transesophageal and transgastric positions, respectively. Two-dimensional images of the left atrium (LA), right atrium, left ventricle (LV), mitral valve, aortic valve, and aorta were recorded.

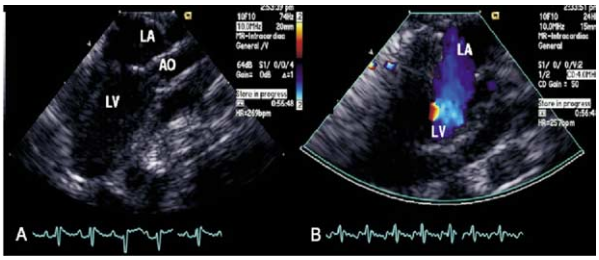

Figure 1 Two-dimensional long-axis view of left ventricle (**A**) and color Doppler (**B**) of mitral inflow obtained from transesophageal study. *LV*, Left ventricle; *LA*, left atrium; *AO*, aorta.

Doppler study. Pulsed wave Doppler and color Doppler studies were performed to assess mitral inflow, pulmonary venous flow, and pulmonary artery flow in the esophageal views. Color M-mode Doppler and Doppler tissue imaging study were also carried out. No angle correction was used for Doppler study because blood flow direction through the mitral valve and ultrasound beam was less than 20 degrees in the LV long-axis view.

M-Mode. After completion of the long-axis view examination of the LV, the catheter was slightly advanced into the rat's stomach about 1 to 2 cm. A transgastric view of the long axis of the LV was obtained and M-mode images were recorded.

Necropsy

Necropsies were performed in 3 rats. The esophagus was incised longitudinally. Visual and pathologic inspections were performed to examine for evidence of esophageal injury.

RESULTS

All attempts at insertion of the ICE catheter in the esophagus were successful. The catheter was usually advanced into the esophagus easily. Initial resistance was encountered in two rats. After repositioning the rats by slightly stretching the rats' necks posteriorly and lifting the intubation tube anteriorly, the ICE catheter was easily reinserted. The images of heart were usually obtained with the catheter tip 8 to 11 cm from the incisors.

Standard Images

Figure 1 illustrates transesophageal long-axis view of the LV. In this view, the LA, the LV, the mitral valve, and the aorta were visualized by 2D echocardiography (Figure 1, *A*), and mitral inflow was evaluated by color Doppler (Figure 1, *B*). This view was also useful for interrogating LV filling by pulsed wave Doppler (Figure 2, *A*) and color M-mode Doppler (Figure 2, *B*). MR could be assessed by color Doppler in the long-axis view. There was no MR detected