IN VITRO ABLATION OF CARDIAC VALVES USING HIGH-INTENSITY FOCUSED ULTRASOUND

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Abstract—The purpose of this study was to evaluate the possibility of using high-intensity focused ultrasound (US), or HIFU, to create lesions in cardiac valves in vitro. Calf mitral valves and aortic valves were examined. Focused US energy was applied with an operating frequency of 4.67 MHz at a nominal acoustic power of 58 W for 0.2, 0.3 and 0.4 s at 4-s intervals. Mitral valve perforation was achieved with 20.8 ± 3.7 exposures of 0.2 s, 15.4 ± 2.1 exposures of 0.3 s or 11.2 ± 2.3 exposures of 0.4 s. Aortic valve perforation was achieved with 13.3 ± 2.4 exposures of 0.2 s, 10.3 ± 2.2 exposures of 0.3 s or 8.4 ± 1.8 exposures of 0.4 s. The mean diameter of the perforated area was 1.09 ± 0.11 mm. The lesions were slightly discolored and coagulation of tissue around the perforation was observed. HIFU was successful in perforating cardiac valves. With further refinement, HIFU may prove useful for valvulotomy or valvuloplasty. (E-mail: otsukaryo@aol.com) © 2005 World Federation for Ultrasound in Medicine & Biology.

Key Words: High-intensity focused ultrasound, Ablation, Cardiac valve.

INTRODUCTION

High-intensity focused ultrasound (US), or HIFU, is a noninvasive extracorporeal technique capable of thermally ablating subsurface structures without injuring intervening tissues (Fry et al. 1955). Ultrasonic energy can be applied in a target volume to induce molecular agitation, absorptive heating and ultimately thermal coagulative tissue necrosis. It is well-known that there are several other thermal ablative techniques, such as argon cryotherapy or interstitial laser therapy (Doll et al. 2003; Gaita et al. 2000; Isner et al. 1985a, 1985b; Kimman et al. 2001, 2003; Selle et al. 1986; Williamson et al. 1993). These techniques cool or heat primarily by thermal conduction and create a graded response depending on the distance from the thermal source. Because this mechanism is relatively slow, it is susceptible to cooling near blood vessels. In contrast, HIFU has an advantage over these techniques, because the tissue in the acoustic focal volume during HIFU ablation is rapidly heated by a remote energy source (the ultrasonic transducer) and the intervening tissue is not damaged.

HIFU is being explored as a therapeutic modality in almost every tissue that is accessible by US. Several studies have examined the histologic changes related to HIFU ablations in the liver, kidney, prostate, breast, brain and myocardial tissues (Damianou 2003; Lee et al. 2000; Malcolm and ter Haar 1996; Sibille et al. 1993; Strickberger et al. 1999; Susani et al. 1993; Vykhodtseva et al. 1994; Wu et al. 2001, 2003; Chen et al. 1999; Yang et al. 1993). However, there are relatively few studies that have examined the possibility of using HIFU for ablation of the cardiac valves, even although several applications could potentially benefit from HIFU.

We hypothesized that HIFU has the potential to reduce the severity of the cardiac valve stenosis by disrupting adhesions. The purpose of this preliminary study was to investigate the effects of HIFU energy on normal cardiac valves and to document the histologic changes induced by it.
MATERIALS AND METHODS

Material

We examined 15 calf mitral valves and 15 aortic valves. We dissected these valves from 15 calf hearts. Each specimen was mounted on a rubber sheet and then placed in a plastic container filled with phosphate-buffered saline (PBS), which was degassed by a deaerator and a dry vacuum pump (model 2560, Welch Ritschle Thomas, Skokie, IL). The container was immersed in a water bath, with temperature maintained at 37.0°C using an electric heater (model 1112A, VWR International, West Chester, PA) (Fig. 1).

High-intensity focused ultrasound

The HIFU energy was supplied with a Sonocare model CST-100 ultrasound therapy system, originally designed for clinical glaucoma therapy. This system consists of a signal generator, a power amplifier and 80-mm spherically shaped bowl HIFU transducer made from piezoelectric ceramic (PZT-4). The transducer’s focal length is 90 mm; it has a central 23-mm hole that houses a 7.5-MHz A-mode diagnostic transducer (Panametrics; Waltham, MA; model MD 3657). The diagnostic transducer is aligned to be coaxial and confocal with the HIFU transducer.

The operating frequency of the HIFU transducer was 4.67 MHz. The focal zone beam shape was measured using a pulse-echo reciprocity technique with a point target; at the half-power points, the focal zone was 4 mm axial and 0.4 mm transverse. The focal zone was also visualized with a Schlieren system (Fig. 2). The acoustic power was 58 W as determined by the radiation force on an absorber. This corresponds to a nominal spatial average intensity of 5 kW/cm².

The transducer assembly was attached to an acrylic resin coupling cone with a 25-mm diameter exit hole. The cone was filled with degassed water and the exit hole was covered with a polyvinylchloride membrane. The focus of the US beam occurred at a depth of 2.5 cm from the tip of the coupling cone; it was positioned at the desired tissue location by distance measurements made with the diagnostic A-mode transducer.

We planned the timing of our exposures based on considerations that this method will be eventually used for a beating heart. For in vivo studies, the exposure timing must be synchronized with the heart rate and HIFU exposures must be made when the cardiac valves are closed. When the heart rate is from 40 to 100 beats per min, the duration of systole or diastole is between 0.2 and 0.4 s. Based on these considerations, the HIFU beam was activated for 0.2, 0.3 or 0.4 s at 4-s intervals. A custom software package was used to control the HIFU system (Fig. 3). We examined five valves for each set of these HIFU exposure parameters.

We produced 10 lesions in each valve. As soon as the perforation was visually affirmed on the target site, HIFU ablation was terminated and we recorded the number of HIFU pulses that had been applied. We measured the diameters of the perforated lesions using an electric slide gauge and computed means and SDs for correlation with exposure parameters.

After HIFU ablation, each leaflet was fixed in 10%
formalin and cut in paraffin blocks. Standard hematoxylin and eosin (H&E)-stained slides and trichrome stained slides were evaluated for pathologic evidence.

**Statistical analysis**

Results are expressed as mean values ± one SD. For each valve, the effect of pulse duration on the number of exposures required for perforation was analyzed using a one-way repeated measures analysis of variance. The number of exposures was assumed to follow a Poisson distribution. For each level of duration, the difference in the number of exposures required for perforation between valves was compared using a paired *t*-test. Statistical significance was defined as a *p* value of less than 0.05.

**RESULTS**

HIFU ablation could be performed on all valves (Fig. 4). Evidence of perforation was visually apparent on all ablated valves (Table 1). The thickness of mitral valves was 0.85 ± 0.28 mm and the thickness of aortic valves was 0.40 ± 0.16 mm. Twelve lesions exhibited discolored areas around the perforated lesions. Mitral valve perforation was achieved with 20.8 ± 3.7 pulses of 0.2-s duration, 15.4 ± 2.1 pulses of 0.3-s duration or 11.2 ± 2.3 pulses of 0.4-s duration (Fig. 5a). Aortic valve perforation was achieved with 13.3 ± 2.4 pulses of 0.2-s duration, 10.3 ± 2.2 pulses of 0.3-s duration, or 8.4 ± 1.8 pulses of 0.4-s duration (Fig. 5b). The required number of pulses with each duration was significantly smaller for aortic valves than for mitral valves (*p* < 0.01). For both valves, the number of required pulses was significantly reduced by increasing the pulse duration (*p* < 0.01). The mean diameter of the perforated lesions was 1.09 ± 0.11 mm, which is approximately 2.5 times larger than the half-power diameter of the focused HIFU beam. There was no significant difference between the mean diameters of the perforations in mitral and aortic valves.

**Pathologic changes**

The valve lesions include central circular defects within a rim of hyperchromatic tissue (Figs. 6 and 7). Prominent pathological findings were a coagulation of collagen tissue around the perforation, which increased stain uptake with a definable border, and the presence of many small vacuoles, thought to be microbubbles, within

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**Table 1. Comparison of the number of pulses and the lesion size in both valves**

<table>
<thead>
<tr>
<th>Pulse duration (s)</th>
<th>Number of required pulses</th>
<th>Perforation diameter (mm)</th>
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<tbody>
<tr>
<td><strong>Mitral valve</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.2</td>
<td>20.8 ± 3.7*</td>
<td>1.09 ± 0.12</td>
</tr>
<tr>
<td>0.3</td>
<td>15.4 ± 2.1*</td>
<td>1.11 ± 0.11</td>
</tr>
<tr>
<td>0.4</td>
<td>11.2 ± 2.3*</td>
<td>1.11 ± 0.11</td>
</tr>
<tr>
<td><strong>Aortic valve</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.2</td>
<td>13.3 ± 2.4</td>
<td>1.08 ± 0.10</td>
</tr>
<tr>
<td>0.3</td>
<td>10.3 ± 2.2</td>
<td>1.09 ± 0.11</td>
</tr>
<tr>
<td>0.4</td>
<td>8.4 ± 1.8</td>
<td>1.11 ± 0.11</td>
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*p* < 0.01 for the comparison with the value for the number of exposures on aortic valve.
the hyperchromatic area. The diameters of these microbubbles were from 10 to 30 μm. In the definable border zone, traces of torsional stress were seen (Fig. 6).

**DISCUSSION AND SUMMARY**

In this study, we found that HIFU could produce ablation of cardiac valves. Creation of perforated lesions in the cardiac valves was very consistent and reproducible.

**Comparison with other techniques**

There are several other thermal ablative techniques, such as carbon dioxide laser, Er:YSGG laser, Ho:YAG laser and radiofrequency (RF) ablation (Goldberg et al. 1998; Huang et al. 1987; Lindsay et al. 1993; Isner et al. 1985a, 1985b; Selle et al. 1986; Williamson et al. 1993).

Previous studies showed that these techniques were effective in the decalcification and debridement of aortic valve leaflets. However, carbon dioxide laser radiation is highly absorbed by water, which constitutes 60% of human tissues, and laser techniques can only be performed when the aortic valve is directly visible. These techniques are susceptible to cooling by nearby blood vessels, because these mechanisms are relatively slow (Kolios et al. 1995). In contrast, HIFU beams can be focused and transmitted through solid tissues within the body to destroy the in-depth lesions, without surgical exposure or insertion of instruments. RF heating and HIFU produce lesions with similar histologic injury in myocardial tissue. However, RF is not focused and the generated energy is absorbed proportionally by the distance between the tissue and the RF catheter. On the other hand, because the HIFU system can focus its energy, smaller and more precise lesions can be created. In addition, HIFU is capable of thermally ablating subsurface structures without a catheter; as such, HIFU ablation can be an alternative technique to the other thermal ablative techniques.

**Pathologic changes and mechanisms**

The perforated valvular holes could be generated accurately and repeatably by HIFU ablation. HIFU is capable of producing lesions not only thermally but also through cavitation, acoustic streaming and shear stresses (Yang et al. 1993; Hill and ter Haar 1995).

We hypothesize that heating is the primary factor in the perforated lesions that we observed. The valves are largely collagenous and previous studies on the thin (0.5 mm) collagenous sclera in rabbits showed that lower...
exposures than those employed in the current study produced a dramatic sclera thinning, immediately rendering it translucent (Coleman et al. 1985). At higher exposures, perforation was observed, as seen in our results. Electron microscopy in the rabbit study showed dissociation of collagen fibers into thin filamentous components. In the current experiments, similar thermal denaturation would lead to thinning and loss of structural integrity.

Mechanical phenomena, mediated by radiation force or streaming (in the coupling fluid adjacent to the valves), may also play a role in the perforations that we observed (Coleman et al. 1985). Evidence for this comes from the signs of torsional stress observed near the perimeters of the perforations. These phenomena could well act synergistically with the thermally weakened collagen fibers to expedite perforation.

We observed small tears and microbubbles around the perforated lesions (hyperchromatic area) in the pathologic examinations. The tears are most likely associated with tensile stress during perforation. The small vacuoles that we observed are most likely associated with microbubbles, which can arise from cavitation and vaporization (boiling of fluids within in tissue) during HIFU exposures (Hill et al. 1995; Bailey et al. 2001). The production of such bubbles can be problematic in many HIFU applications, because they amplify the attenuation of US and block US passage to posterior regions, shielding them from the therapy beam. However, such considerations do not pose significant problems for the current application; the bubbles which we observed were peripheral to the treatment area and are not important, because of the small thicknesses of the valves.

It is interesting to note that perforations of the same diameter (1.09 ± 0.11 mm) were produced in both types of valves. In mitral valves, these were produced by pulses with a total “on” time (product of number of required pulses and pulse duration) of 4.36 s (mean) with a range of only ± 0.3 s. The range is only ± 7% of the mean, in spite of the two-to-one range in pulse duration. The same type of observation holds true for aortic valve results, except that the mean total “on” time is 3.37 s with a range of 0.3 s (8% of the mean). These results imply that the perforations are produced when the incident energy has reached a critical level. The results suggest that perforations can be produced when \( w \ DTP > E \), where \( w \) is the incident acoustic power, \( D \) is pulse duration, \( T \) is the total exposure time, \( P \) is the pulse repetition rate (PRR) (pulses/s) and \( E \) is a valve-specific energy threshold. Thus, all other factors being constant, if the interpulse period can be shortened to 1 s (\( p = 1/s \)), the required total exposure time can be reduced by a factor of four from the values in this report (\( p = 0.25/s \)).

Clinical implications

Elderly patients with severe symptomatic aortic stenosis are treated poorly with medical management (Hufnagel and Conrad 1962; Roberts et al. 1971). The optimal treatment for these patients is surgical valve replacement (Kirklin and Mankin 1960). Noninvasive valvuloplasty may be useful as a bridge to aortic valve replacement in hemodynamically unstable patients, in patients undergoing emergency noncardiac surgery and in patients with comorbid diseases that are too severe to undergo cardiac surgery. There has been a great interest in aortic and mitral balloon valvuloplasties (Rahimtoola 1987; Otto et al. 1994). However, these techniques are invasive and have limited effectiveness (Stoddard et al. 1989).

Further investigation

In this study, we employed a HIFU transducer whose focused beam has a circular cross-section, as defined by its half-power points. We plan to study other transducers with asymmetrical strip electrodes that produce focused beams confined within elliptical rather than circular focal-plane regions (Lizzi et al. 1998).

We plan to study whether or not these beams can produce narrow incisures. Such shapes may be more effective in cardiac valve ablation.

We will investigate HIFU ablation on calcified valves in vitro as the next study. US energy is strongly absorbed and attenuated by calcified tissues. Therefore, the ablation of calcified valves may require a larger number of exposures or the use of higher intensities.

In the future, we will examine valve ablation in in vivo animal models. These studies will employ expanded versions of hardware and software employed in the current study. The current system software provides control over the duration and repetition rate used in exposure and it already incorporates electrocardiogram gating to synchronize exposures with the cardiac cycle.

CONCLUSIONS

Although the results of this study are preliminary, they demonstrate that HIFU can create superficial thermal lesions and perforation in mitral valve and aortic valve tissues. This technique may lead to an US-based valvuloplasty.

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REFERENCES


