Cardiovascular Damage after cw and Q-switched 2µm Laser irradiation

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ABSTRACT

Aiming for laser-assisted resection of calcified aortic valve structures for Transcatheter Aortic Valve Implantation (TAVI), a Q-switched Tm:YAG laser emitting at a wavelength of 2.01 µm was used to evaluate the cutting efficiency on highly calcified human aortic leaflets in-vitro. The calcified aortic leaflets were examined regarding ablation rates and debris generation, using a pulse energy of 4.3 mJ, a pulse duration of 0.8-1 µs and a repetition rate of 1 kHz. The radiation was transmitted via a 200 µm core diameter quartz fiber. Resection was performed in a fiber-tissue contact mode on water-covered samples in a dish. The remnant particles were analyzed with respect to quantity and size by light microscopy. Additionally, soft tissue of porcine aortic vessels was examined for histologically detectable thermo-mechanical damage after continuous wave and Q-switched 2µm laser irradiation.

An ablation rate of 36.7 ± 25.3 mg/min could be realised on highly calcified aortic leaflets, with 85.4% of the remnant particles being <6 µm in diameter. The maximum damaged area of the soft tissue was < 1 mm for both, cw and pulsed laser irradiation. This limits the expected collateral damage of healthy tissue during the medical procedure. Overall, the Q-switched Tm:YAG laser system showed promising results in cutting calcified aortic valves, transmitting sufficient energy through a small flexible fibre.

Keywords: calcified aortic valve; in vitro; debris; laser resection; Q-switch laser

1. INTRODUCTION

In recent years, minimal invasive surgery was developed for replacing artificial heart valves to avoid open heart surgery. In existing methods, these pathological - usually calcified - valves remain in the body and are only pushed aside by the artificial valve [1]. This may lead to deformation of the artificial valve and therefore to leakage and higher wear [2]. In order to avoid this, a minimally invasive method to resect the original valves was proposed and demonstrated in animal trials using a cw 2µm laser system [3].

Unlike porcine heart valves, pathological human valves are calcified, consisting of a mixture of hard and soft tissue, demanding a different laser system for efficient cutting. To meet this requirement we designed a Q-switched, high repetition rate laser system at a wavelength of 2 µm, which is capable of delivering the energy via optical fibre for minimal invasive surgery. The cutting properties, sizes of the generated debris, as well as the histological damage to surrounding soft tissue, are studied.

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2. MATERIAL AND METHOD

2.1 Heart valve samples
Calcified aortic valves were obtained from patients (N = 11) undergoing planned aortic valve replacement (approved by the Ethics Committee of the Christian—Albrechts—University of Kiel from the 24th of November 2004 (D 434/04)). Only highly calcified aortic valves were used for the experiments. The specimens were stored in a 0.9% solution of sodium chloride until the experiments were performed.

2.2 Laser System
An experimental diode pumped Tm:YAG laser setup was developed. An acoustic-optic modulator (Gooch & Housego, Ilminster, UK) was used as a Q-switch to generate pulses with a duration of 0.8 to 1 µs. This system emits pulsed laser radiation at a wavelength of 2013 nm with an average pulse energy of 4.3 mJ at a repetition rate of 1 kHz.

The radiation was transmitted via a 200 µm core diameter low OH fiber with a numerical aperture of 0.22 (All Silica Low OH CF01493-51, Laser Components, Olching, Germany). This corresponds to a radiant exposure of 12.7 J/cm² per pulse and an intensity of 15.2 MW/cm² at the distal fiber tip, assuming a lateral top hat energy distribution.

2.3 Resection procedure
In order to quantify the cutting efficiency calcified human aorta leaflets were cut in a dish under water, mimicking the water filled resection chamber for the medical procedure. The cutting efficiency is measured by the loss of mass before and after the procedure for each sample. The fiber was mounted in a steel tube as a handpiece and the radiation was applied in fiber-tissue contact mode. After the first cut within a time period of one minute, the water in the Petri dish was filtrated with a 0.4 µm pore diameter filter sheet (Millipore XX1004704 filter system and Millipore IsoporeTM Membranfilter, Millipore GmbH Germany). Subsequently, two additional cuts were applied. Thus a total of three cuts with a duration of one minute each were applied to every sample to minimize the relative measurement error, leading to an overall exposure of three minutes.

2.4 Debris analysis
After drying of the membrane filter, pictures were taken with a microscope (Leitz Wetzlar Orthoplan, Wetzlar, Germany) and a camera (Leica DC 300, Wetzlar, Germany) at two different magnifications of 2.52 and 6.3. Then the particles cross section and quantities were calculated with the open source software package ImageJ (version 1.42q, Wayne Rasband, National Institute of Health, USA).

30 sections of each membrane were photographed with a magnification of 2.52 in order to analyse the particle sizes of 10 to >300 microns. This corresponds to 18.7 % of the surface area of the filter with a pixel edge length of 1.4 µm. Additional 28 sections were taken at a magnification of 6.3 thus covering 2.2 % of the surface with a pixel edge length of 0.5 µm to analyse particle diameters of < 2 to 40 microns. The sections were aligned in a cross shaped pattern over the filter area in order to avoid an overlap of the analyzed domains.

ImageJ was used to identify and measure the particles by setting a gray scale threshold to identify the particles and determine the size and quantity for different diameter intervals. An upper and lower greyscale threshold was set manually to mark the particles in the picture. The area A of each particle was automatically calculated by the software and an equivalent circular diameter was calculated according to $d = 2 \times \sqrt{A / \pi}$.

The quantities measured at 58 random locations were extrapolated to the full filter area. The difference between the maximum length of the irregular formed particles and the idealised diameter was inspected for some arbitrarily chosen particles. It was found that the maximum length of some particles differ up to 25 % from the idealised circle diameter.
2.5 Histological micrographs
To estimate the mechanical and thermal damage to soft tissue, porcine aorta samples were placed under saline solution and irradiated with the fibre tip in contact with the tissue. To ensure a uniform pulse delivery to the samples, a linear stage was used under an angle of 45 degree and a feed of 120 mm per minute. Afterwards the samples were histologically analysed.

3. RESULTS

3.1 Ablation rate
The averaged ablation rate was 36.7 ± 25.3 mg per minute on highly calcified aortic leaflets. The cuts were carried out over the hard tissue in the aortic leaflets on eleven samples. Employing this technique, calcified aortic leaflets could be cut in about 3 minutes.

![Ablation Rate Graph](image)

Figure 1: Ablation rate different samples, the error bars denote the standard derivation. Resection time was 3 times one minute, pulse parameters are given in the text. The solid line represents the average ablation rate of 36.7 ± 25.3 mg/min.

3.2 Debris analysis
Figure 6 shows the quantity of particles in a logarithmic scale for different diameter intervals of the debris. In general, this graph shows that particle numbers are decreasing for increasing particle diameters and that the majority of particles is smaller than 2 µm. The overall quantity of particles is 2.25*10⁶, while 1.3*10⁶ particles are smaller than 2 µm in diameter, which corresponds to a fraction of 59%. Only 17.5 % of the particles appear in the following diameter interval between 2 and 4 µm and 8.9 % between 4 and 6 µm. Accordingly a majority of 85.4 % of all particles are found to be smaller than 6 µm. For particle diameters larger than 100 µm, the relative fractions of the particle numbers are below 1%, the largest observed diameter was 800µm.
3.3 Histological micrographs

The mechanical and thermal damage of Q-switched and cw laser radiation to soft tissue histological micrographs are shown in Fig 3. The laser parameters and histological damage zones are listed in Table 1. Both types of radiation show comparable thermal damage zones well below one millimetre. The bridge remaining on the top side of the left sample is due to bending of the fiber while moving across the not perfectly flat sample. In this way the fibre shortly points sideways at an lower angle, thus creating a hole and not a cut.

Figure 3: Histological micrographs of porcine aorta irradiated with Q-switched (left) and cw (right) 2µm laser radiation.
Table 1: cutting depth and width of porcine tissue for different laser parameters.

<table>
<thead>
<tr>
<th>Fig.</th>
<th>Tissue</th>
<th>Mode of operation</th>
<th>zone depth [µm]</th>
<th>zone width [µm]</th>
</tr>
</thead>
<tbody>
<tr>
<td>3 left</td>
<td>Porcine</td>
<td>Q-switched, 1kHz</td>
<td>174 (red)</td>
<td>215 (red)</td>
</tr>
<tr>
<td></td>
<td>Aorta</td>
<td>4.5±0.2 mJ (max. 5.1 mJ)</td>
<td>241 (orange)</td>
<td>358 (orange)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>337 (green)</td>
<td>708 (green)</td>
</tr>
<tr>
<td>3 right</td>
<td>Porcine</td>
<td>cw 4.3 W</td>
<td>142 (red)</td>
<td>243 (red)</td>
</tr>
<tr>
<td></td>
<td>Aorta</td>
<td></td>
<td>225 (orange)</td>
<td>442 (orange)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>359 (green)</td>
<td>798 (green)</td>
</tr>
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</table>

4. DISCUSSION

4.1 Ablation rate

This study shows that calcified aortic leaflets can easily and fast be ablated by the pulsed, high repetition rate 2µm laser radiation. The ablation rate was 36.7 ± 25.3 mg per minute. This is a significant improvement compared to 6.7±26.0 mg per minute shown in an earlier study with cw laser radiation of 25 W, which failed to cut strongly calcified sections of the samples [4].

In most cases it was possible to cut through an aortic valve leaflet within three minutes. Therefore, it is expected to take about 9 minutes of laser procedure for a resection of a complete valve with three leaflets.

The obvious advantage of the 2 µm laser wavelength is due to the absorption of the resection media. In soft tissue, a high water absorption leads to strong local heating and therefore strong thermal effects. Calcification however cannot be broken by thermal effects, thus the resection effect is reduced. The high intensity of pulsed laser radiation however leads to photomechanical and photodisruptive effects which are capable of resecting calcified tissue.

4.2 Debris

For the particle analysis the remnant particles were filtered, photographed and evaluated. The accuracy of the developed method for debris analysis is limited by different factors as pixel size, manual setting of upper and lower greyscale thresholds and shading of filter and particles. In order to minimise the influence of these errors a statistic over 11 samples with 58 microscopic pictures each was used. A comparison of the maximum length of the irregular shaped particles to the idealised circular diameters on arbitrarily chosen particles shows a difference up to 25 %.

The particle analysis showed a wide distribution of different particle diameters with a fraction of 85% of the particles being smaller than 6 µm in diameter, which is less than the size of erythrocytes with a diameter of 7 µm. Less than 1 % of the analysed particles are larger than 100 µm in diameter, which corresponds to several hundred of these large particles ablated in one minute. Since a complete resection is expected to take about 9 minutes, accordingly more debris is expected.

To inhibit the particles entering the human vascular system and avoid risk of thrombosis for the patient, an isolated resection chamber is needed, which prevents the escape of debris during the resection process as described in [5]. The requirement for the resection chamber is to seal the encapsulated volume without leakage of small particles, and a drain pipe with an inner diameter of about 1 mm appears reasonable to allow rinsing of the chamber. The resection of native tissue has recently been demonstrated in a beating heart [3]. The laser investigated in this study together with the isolation technology show a very promising approach to resect calcified valves to come closer to the cardiac surgical golden standard.
4.3 Histological damage

Furthermore the histological damage to soft tissue surrounding the aortic leaflets is in the same order of magnitude for both, Q-switched and cw laser radiation and well below 1 mm. The Q-switched pulsed treatment showed no stronger ruptures of the soft tissue, thus only moderate collateral damage is expected during medical procedure. This has to be taken into account, because it is possible for the fiber to get into contact with healthy aorta tissue surrounding the pathological valves.

5. CONCLUSION

In conclusion, the pulsed 2µm laser system is a promising dissection device for percutaneous aortic valve replacement. In order to increase the resection rate with this laser system, the investigation of different pulse parameters and the increase of the pulse energy appear reasonable. Further research and development are needed before first clinical trials can be initiated, allowing a catheter based removal of the aortic valve. However, clinicians already recognized the need for an improved TAVI technology if this were to be applied to younger patients.

ACKNOWLEDGMENTS

This project was realized with the support of the Deutsche Forschungsgemeinschaft (Br 1349/2-1).

REFERENCES