Management of Heat in Laser Tissue Welding Using NIR **Cover Window Material**

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Background: Laser tissue welding (LTW) is a novel method of surgical wound closure by the use of laser radiation to induce fusion of the biological tissues. Molecular dynamics associated with LTW is a result of thermal and non-thermal mechanisms.

Objectives: This research focuses exclusively on better heat management to reduce thermal damage of tissues in LTW using a near infrared laser radiation.

Methods: An infrared continuous-wave (CW) laser radiation at 1,450 nm wavelength corresponding to the absorption band from combination vibrational modes of water is used to weld together ex vivo porcine aorta.

Results: In these studies we measured the optimal laser power and scan speed, for better tensile strength of the weld and lesser tissue dehydration. Significant amount of water loss from the welded tissue results in cellular death and tissue buckling. Various thermally conductive optical cover windows were used as heat sinks to reduce thermal effects during LTW for the dissipation of the heat. The optimal use of the method prevents tissue buckling and minimizes the water loss. Diamond, sapphire, BK7, fused silica, and IR quartz transparent optical cover windows were tested.

Conclusions: The data from this study suggests that IR-quartz as the material with optimal thermal conductivity is ideal for laser welding of the porcine aorta. Lasers Surg. Med. © 2011 Wiley Periodicals, Inc.

Key words: laser tissue welding; heat management; aorta; water absorption; CW 1,450 nm fiber laser

INTRODUCTION

The use of laser energy to fuse well opposed tissue segments has been applied to tissues from different organ sites including: vascular [1-3], skin [3-5], ocular [6,7], urology [8], and gastrointestinal tract [9-11]. This technique called laser tissue welding (LTW), provides an effective and rapid means of water-tight wound closure [3,12,13] with the potential for reduced scar formation [14,15]. There are currently two approaches to LTW. The first approach employs lasers which emit at wavelengths which are absorbed by native absorbers found in tissue, such as the water absorption band. The second approach uses extrinsic dyes or proteins as the absorbers, and is more akin to laser soldering. LTW using native tissue absorbers reduces the risk of infection or foreign body reactions. Much of the LTW research using native tissue absorbers has focused on using either CO₂ lasers [16–18], or in Alfano's group near infrared (NIR) lasers tuned to vibrational combination modes of water around 1,455 nm [3,5,7].

Although the exact mechanisms of LTW are not well understood, in NIR LTW, the laser energy is absorbed by water which is naturally present in tissue and then transferred to the collagen triple helix. The vibrational modes of water becomes hot to transfer energy to collagen. The relatively moderate strength of water absorption at 1,455 nm (absorption coefficient of water at 1,455 nm is 31 cm⁻¹) allows full thickness welding in tissues with a depth of up to 2 mm. LTW at 1,455 nm permits significantly deeper penetration then with CO_2 lasers at 10.6 μ (penetration depth in tissue is $10-20 \ \mu m$ at $10.6 \ \mu m$). Weld depth can be further controlled by tuning the welding laser away from the absorption peak [3,5,7,13]. NIR LTW in the 1,455 nm region has been applied to ex vivo human and porcine aorta [3], skin [3], and ocular [7] tissues. In NIR LTW, the tissue is heated to temperatures slightly greater than 60° C. The heating disrupts and partially dissociates collagen bonds. Subsequent cooling restores both covalent and non-covalent bonds [1,19–23], and reforms the collagen helix. This re-naturation of the collagen helix binds the tissue across the incision with a water tight seal which is indistinguishable from nonwelded tissues under histopathologic or electron microscopic examination [3,7]. The collagen helices are bound together with both hydrogen and water mediated bonds, therefore hydration is important for stabilizing the interactions between the collagen helices [24,25] and maintaining tissue hydration is important to insure high quality welds. The relative contributions of free and bound water in LTW are not presently known. Precise control of the tissue temperature through modulation of laser power, exposure time, and illumination area is essential for successful welding [3,7,26]. It is crucial to maintain the hydration level of the tissue during LTW [27].

Conflict of interest: none.

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Although LTW has important advantages over conventional methods of wound closure, several issues need to be addressed in order to move LTW into clinical practice. Some potential problematic areas in LTW are: (i) inconsistent weld strength-while most welds have high tensile strength, some tissues fail to bond; (ii) risk of thermal damage-using too high a laser power or too long an exposure time can result in thermal damage; (iii) tissue dehydration-water loss during welding dehydrates the tissues and may limit weld effectiveness; and (iv) tissue buckling-thermal expansion of the tissues during welding may cause the two tissue pieces to separate. To reduce thermal damage, welding was performed with a highspeed (5 mm/s) scanning laser using a pattern consisting of multiple lines. The lines had varying offset from the incision line to distribute the laser energy over a larger area. The use of ps and fs light pulses further reduces thermal damage effect and less scaring.

This work reports, the effects of using different transparent cover windows for heat management of laser welds of ex vivo porcine aorta tissues. Transparent cover windows were positioned over the tissue to reduce water evaporation, prevent buckling, and to serve as heat sinks to cool the tissues from NIR laser radiation. We investigated LTW using cover window materials of different thermal conductivities to maximize tensile strengths.

MATERIALS AND METHODS

Sample Preparation

All the animal experiments done in this study were approved by IACUC of the City College of New York. The porcine aortas were purchased from a USDA approved animal slaughter house (Countryside Quality Meats Inc, Union City, MI). The porcine tissues were shipped to our facility overnight on wet ice. The received samples were washed and cleaned with 0.9% saline solution. The aortas were filleted open by an incision along their long axis and then cut into rectangular pieces of size $20 \times 100 \text{ mm}^2$, with the long axis of the rectangular pieces parallel to the direction of blood flow. These pieces were then sectioned into $20 \times 10 \text{ mm}^2$ rectangular pieces with the longer side being parallel to the circumference of the aorta, as shown in Figure 1. An incision was then made in the $20 \times 10 \text{ mm}^2$ to create two $10 \times 10 \text{ mm}^2$ pieces which are

then reapproximated and welded back together. In this geometry, the incision is parallel to the direction of blood flow and most of the collagen fibers are perpendicular to the incision. Samples' weights were measured before and after welding with the weight difference being due to water loss.

Cover Windows

Two cover windows were placed over each tissue to be welded (except for the control specimens which had no cover windows) with a gap along the incision line. The tissues were also mounted on a glass slide. The cover window materials were: 1 mm thickness diamond, sapphire, fused silica, quartz, and BK7 glass. The cover windows were transparent at 1,455 nm and had thermal conductivities which varied from a low of 0.01 (BK7 glass) to a high of 6.1 W/cm K (diamond). The thermal conductivity of human aorta tissue is about 0.0048 W/cm K [28]. Thermal conductivity of porcine aorta should be close to that of human aorta.

Laser System

LTW was performed with a continuous wave (CW) erbium fiber laser, B&Wtek model BWF2 (B&W Tek, Inc., Newark, Delaware), emitting at 1,455 nm. The power level of the laser is user adjustable with a maximum output of 2 W. All specimens were welded at a power of 900 mW. The laser light was delivered to the tissue by an optical fiber and focusing lens. The laser spot size at the focal plane was 80 μ m in air. When tissue samples were present, scattering significantly increased the focal spot size. Specimens were positioned such that the focal plane was approximately half way into the tissue. The BWF2 is equipped with an electronic shutter which was computer controlled to allow precise control of the laser exposure time. A red LED included in the BWF2 was used for precise alignment of the tissues relative to the laser beam.

Translation System

The porcine aorta samples were fixed on a computercontrolled two axes translation stage (see Fig. 2). Samples



Fig. 1. The method for ex vivo porcine aorta sample preparation for LTW.



Fig. 2. Schematic diagram of the LTW system.

were positioned such that the incision was parallel to the x-axis stage. Translating the tissues along the x-axis permitted the laser to weld along the entire 10 mm incision and provided time for the tissue to cool between scans. Motion along the y-axis permitted the sample to be offset relative to the laser, distributing the laser energy over a larger area. This permitted the use of higher laser power and resulted in shorter welding times, without increasing thermal damage. The tissues were welded using a system of 56 patterns. Each pattern consisted of eight lines with different y-axis offsets relative to the incision. Four of the eight lines were directly on the incision, two of the lines were offset $\pm 35 \,\mu$ m, and the other two lines were offset $\pm 70 \ \mu m$ relative to the incision. Figure 2 shows a block diagram of the LTW apparatus. The line pattern is shown in Figure 3, with the relative distribution of laser energy in the y-direction, assuming 80 µm spot size, shown at the top of the figure.

Tensile Strength Measurements

After welding, the tensile strength of each of the porcine aorta was measured using a digital force gauge (Model EG025, Mark-10, Hicksville, NY). To determine the tensile strength, the breaking force recorded by the gauge was divided by the surface area of the weld as calculated by the product of the length and thickness of the incision.

Histology

The ex vivo porcine aorta samples from each group that is welded without heat sink and with heat sinks were evaluated histologically to study the extent of changes due to thermal damage during welding. Porcine aorta samples after the welding were immediately transferred to 4% buffered formaldehyde solution for the fixation for 4 days. The tissues then were routinely processed and cut 8 μ thick sections and stained by H&E stain and cover slipped.

RESULTS AND DISCUSSION

The ex vivo porcine aorta tissues were divided into six groups, and each group was welded with the five cover windows materials listed above and a control group without a cover window. Each group consisted of about 20 specimens. Table 1 summarizes the mean tensile strength and fractional water loss of the six groups. The strongest welds were achieved with IR quartz cover windows, fused silica windows yielded next strongest. Both the lower



Fig. 3. The schematic representation of eight line pattern used for pumping NIR laser.

thermal conductivity windows (BK7 glass and no windows) and the higher thermal conductivity windows (diamond and sapphire) resulted in weaker welds. The weld strength plotted as a function of window conductivity is shown in Figure 4. The best tensile strengths were obtained by IR quartz and fused silica windows while the ones with diamond windows did not weld as good. This suggests that very high thermal conductivities of substrates like diamond will not help in LTW as a result of excessive heat dissipation. However, the moderate thermal conductivities of substrates like IR quartz and fused silica helped to enhance the weld quality and tensile strengths significantly. Water loss was minimal with sapphire and IR quartz windows and still yield relatively better weld strengths (see Table 1 and Fig. 5).

The histological evaluation of the laser welded tissue samples revealed very high thermal damage in the porcine aorta tissues welded without any heat sink (Fig. 6A and B). These tissues show extreme levels of tissue shrinkage and cytoplasmic vacuolation as a result of laser generated heat. The tissues welded using IR quartz windows had the best weld strengths and significantly lesser thermal damage was observed in the histological sections (Fig. 7A and B). The next best tensile strengths were obtained using fused silica and sapphire windows however; histologically these tissues show significantly more thermal damage than IR quartz heat sink welds (Fig. 8A and B). Very poor

TABLE 1. Tissue Welding Measurements and Parameters

Tissue type	Number of samples	Cover window	Mean tensile strength (kg/cm ²)	Mean water loss (gm)	Thermal conductivity (W/cm-K)
Pig Aorta	40	None	0.86 ± 0.58	0.034	—
Pig Aorta	27	BK7 Glass	0.73 ± 0.32	0.034	0.0111
Pig Aorta	20	IR Quartz	1.41 ± 0.66	0.021	0.0726
Pig Aorta	10	Fused silica	1.23 ± 0.46	0.055	0.106
Pig Aorta	25	Sapphire	0.95 ± 0.62	0.014	0.33
Pig Aorta	15	Diamond	0.83 ± 0.35	0.030	6.1



Fig. 4. Mean tensile strength of tissue welded with windows of different thermal conductivity.

tissue quality tissue welds were obtained by the use of diamond and Pyrex heat sinks. Both the material left extensive tissue damage and welds were not full thickness (Fig. 9A and B).

Previous studies from our laboratory [3] showed minimal thermal damage to porcine aortic tissue when low CW NIR laser power (approximately 300 mW) was used. However this method took much longer time for each weld. In our current studies, we increased the power of laser up to 900 mW to shorten weld time and to decrease thermal damage we increased the speed up to 8 mm/second. The histological evaluations of the tissues showed increasing the speed alone at high laser energies did not completely prevent thermal damage (Fig. 6A and B). High degree of cytoplasmic vacuolation with accompanying cellular damage confined upper layers of cells which are



Fig. 5. Number of welded and failure samples for different windows.





Fig. 6. H&E stained of porcine aorta welded without any heat sinks. A and B: are at 4X and 10X magnifications, respectively. The tissues show extreme thermal damage.

subjected to intense heat generated by the exposure to laser irradiation. Significantly less cellular damage occurred in deeper layers of cells of welded porcine aortas. The higher temperatures caused extreme dehydration of tissues thus altering the cellular morphology and tissue architecture. Under in vivo conditions such extreme dehydration of tissues may significantly affect healing process; therefore it is essential to find the ways to prevent dehydration. Studies by Fried and Walsh [4] used short 20-100 ms spurts of cryogen to cool tissue surfaces during LTW. Though the results from this study were encouraging, the method has inherent drawbacks such as timing of spurts of coolant gases in relation to climatic conditions like humidity and temperature that exist during the tissue welding and environmental regulations regarding the use of chlorofluorocarbons.

The other researchers have addressed the thermal damage of the tissues during laser welding by incorporation of dyes like indocyanine green [27], India ink [4], and albumin solder [8]. The major drawbacks of these methods included foreign body reactions, inflammation, pigmentation, and local cellular toxicities as result of degradation products of dyes and solder proteins or combinations of both.



Fig. 7. H&E stained sections of porcine aorta welded with quartz heat sinks. **A** and **B**: are at 4X and 10 X magnifications, respectively. The tissue show relatively less thermal damage.

Considering the existing limitations of cooling systems and dyes, for laser tissue surgical procedures in this study we used safe, predictable, reliable cost effective, and environmentally acceptable transparent heat sinks made of materials like IR quartz and Fused silica that produced best welds. The results of tensile strength measurements and histological evaluations of laser welded samples from this study show that weld qualities depend on thermal conductivities of heat sinks. Neither the very high thermal conductivity of diamond nor lower thermal conductivity of BK7's is beneficial for tissue welding procedures using NIR laser in water absorption band range. Rapid dissipation of the heat with use of highly conductive diamond sinks might possibly have prevented the tissue from reaching optimal weld temperature in deeper cell layers resulting in partial thickness welds. Still diamond heat sinks caused rapid high temperatures locally in superficial layers of tissue causing thermal damage as the synthetic diamonds have black inclusions which possibly absorbed the laser radiation apart from dissipating the heat. Therefore, it is essential to have totally non-absorbing material for heat sinks to minimize the thermal damage. The best quality and higher success rates were obtained for welds that used IR quartz heat sinks with a thermal conductivity of





Fig. 8. H&E stained sections of porcine aorta welded with fused silica (\mathbf{A}) magnification 10X and sapphire (\mathbf{B}) magnification 4X Both the sections showing significant thermal damage.

0.0726 W/cm K. The heat sinks made of fused silica and sapphire yielded good tensile strengths and weld quality next to IR quartz. The BK 7 yielded low weld strengths and higher dehydration of tissues due to thermal damage that might have resulted due to its low thermal conductivity.

CONCLUSION

This primary study suggests the need to control thermal damage and to prevent the loss of water from the tissues during LTW to achieve better weld quality and higher tensile strengths using NIR CW laser at 1,450 nm wavelength. The use of ultrafast ps/fs pulse laser radiation around 1,400 to 1,600 nm will be more suitable to reduce thermal effects and tissue burning, which will be the focus of our future work [29]. The use of IMRA 1,535 nm fiber femtosecond laser shows less scaring in animals [30]. The much higher thermal conductivities of substrates like diamond (6.1 W/cm K) will result in too much cooling of the tissues causing total weld failure. Poor thermal conductivities of heat sinks made of substrates like BK7 glass (0.0111 W/cm K) caused same amount of mean water loss from the tissues as seen in the welds without any heat





Fig. 9. High magnification H&E stained sections of porcine aorta welded with diamond (\mathbf{A}) and Pyrex (\mathbf{B}) heat sinks (both at magnification 10X). Both the sections showing thermal damage and partial thickness weld.

sinks. The best results in LTW were obtained with heat sinks with relatively moderate thermal conductivities in the range of 0.07–0.33 W/cm K. The best tensile strengths were obtained by using IR quartz (0.07 W/cm K) followed by fused silica (0.106 W/cm K) and Sapphire (0.33 W/cm K). The speed of laser welding needs to be increased for clinical use in plastic and robotic surgery.

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