



Human Skin Wound Welding Using 980 nm Diode Laser: an *in Vitro* Experimental Study

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Abstract: Laser assisted skin wound closure offers many distinct advantages over conventional closure techniques. The objective of this *in vitro* experimental study, carried out at the Institute of Laser for Postgraduate Studies/Baghdad University, was to determine the effectiveness of 980 nm diode laser in welding of human skin wounds. Multiple 3-4 cm long full thickness incisions in a specimen of human skin obtained from the discarded panniculus of an Abdominoplasty operation were tried to be laser welded using a 4 mm spot diameter laser beam from a 980 nm diode laser at different laser parameters and modes of action. The tensile strength at the weld site was analyzed experimentally. Although laser assisted wound welding did actually happen at many laser parameter settings, it was not strong enough to be clinically useful in living human beings. Despite the 980-nm diode laser system has proved to be an effective way of wound welding but it the weld was of low tensile strength to withstand living patient normal daily activity and movement. It can really shorten the operative time required to close a wound if proper laser parameters were used among an endless effective combinations of parameters. Further future studies are highly recommended on live human beings.

Introduction

Surgery is the science and art of cutting and joining. Conventional methods are currently used for wound edge joining such as surgical sutures, adhesive tape, staples, clips, tissue glue and adhesives. Recently, laser skin welding has been added and it's claimed to be faster, watertight, avoids a foreign-body reaction, minimal inflammation and tissue trauma, and consequently reduced scar formation and improved cosmetic results (Bassam 2009, Wider 1991, Charles 2007). It may even lead to a faster healing since laser energy used may also actually stimulate the tissue healing process (Lon 1999). This technique is becoming

important in many surgical specialties, including urology, cardiothoracic surgery, plastic surgery, and neurosurgery. The method is used especially in instances when stapling or suturing is difficult. However, collateral thermal injury, laser device cost and variability of results has prevented its wide clinical use until now. In experimental animal models, it showed good results but there is paucity of studies that has been published on use of this modality in human beings (Abergel 1986, Fried 2000, Mia 2001).

A 980-nm diode laser has been chosen in this study because it has the advantage of emitting a laser that has a relatively high absorption peak by water and hemoglobin while the absorption coefficient of melanin for 980-nm laser is less

than the absorption coefficients of it for 780- and 815-nm diode lasers leading to even more penetration of laser energy to deeper skin layers (about 3-4 mm) (Murat 2006).

The objective of this *ex vivo* study is to evaluate the effectiveness of 980nm diode laser in laser human skin wounds welding experimentally and to determine whether it would be of a sufficient initial tensile strength to be more applicable clinically in future in vivo studies.

Materials

All procedures and experiments have been conducted at the Institute of Laser for Postgraduate Studies during the period from 1 /7/ 2010 to 1/3/2011.

Human Skin Samples Preparation

In this in vitro study, human skin, harvested from the surgically excised panniculus (Figure 1) of a 45 years old woman who have undergone an Abdominoplasty procedure at Al-Kadhimiyyah teaching Hospital, was used after taking patient's permission. The freshly excised panniculus has undergone a meticulous washing with normal saline then just a layer of 2-3 cm subcutaneous fat was left under the skin. The skin samples were then kept in covered plastic containers in a refrigerator at 4 degree Celsius. Just prior to laser welding experiments, the skin samples were allowed to warm up again slowly to room temperature.

Figure 1 shows a photo plate of part of the excised panniculus composed of human skin and subcutaneous fat.



Fig. (1): A photo plate of the excised human Skin and subcutaneous fat.

The Laser Devices Used

The technical specifications of the chosen 980 nm diode laser device are shown in Table 1. The output laser of this device was delivered to the target site via a 600 micrometer optical fiber. The tip of this optical fiber was kept about 2 mm above the target site during all applications and the spot size of the laser beam has been fixed at 4 mm in diameter. Variable Laser settings and parameters of this laser device were used in this work.

Table (1): VELAS 60, 980 nm diode laser specifications

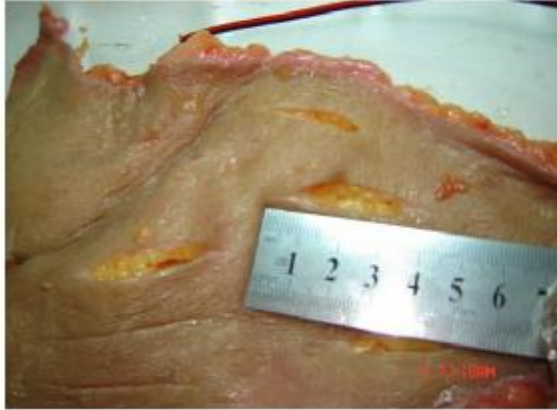
Laser Type	GaAlAs Diode Laser
Model	VeLas60A/B
Wavelength	980nm
Maximum Power	60W
Operation Mode	CW, Single Or Repeat Pulse
Pulse Duration	20-5000ms
Repetition Rate	0.5-20 Hz
Transmission System	600um With SMA905 Connector

Methods

Multiple 0.5-1 cm deep 3-4 cm long full thickness skin incisions were made in the human skin sample, exposing the underlying subcutaneous fat layer (Figure 2).



(A)



(B)

Fig. (2): Multiple 3-4 cm long full thickness skin incisions (B) made in human skin sample (A).

The skin incision edges then were temporarily approximated using dental tweezers (Figure 3) while a 4 mm spot diameter laser was irradiating the incision:



(A)



(B)

Fig. (3): Approximation of the skin incision edges prior to laser irradiation using the dental tweezers

Continuous (at 4mm/sec. speed back and forth laser beam scanning movement), single pulsed spot/spot and repetitive pulse mode laser with scanning movement at variable laser parameters have been applied to the target tissues. The end point of welding of the skin wound was confirmed by naked eye appearance only as swelling and puckering of the wound edges and the wound edge fusion (Figure 4). After letting the skin incision to cool down back to normal ambient temperature again as confirmed by the non contact infrared thermometer, a gentle force (Figure 5) was applied to the wound edges trying to separate them, noticing whether the welding was of a clinically significant strength or not, in other words, is this weld strong enough to withstand the expected normal movements of a live patient in future studies or not.



Fig. (4): A successfully laser welded skin incision with swelling of the wound edges following laser irradiation.



Fig. (5): Application of gentle force trying to separate the laser irradiated wound edges.

Any successful laser welding experiment was repeated 3 times at same laser parameters setting to confirm the reproducibility of the result.

Results

A successful, though weak, skin incision welding has been obtained using the laser parameters and modes of action in table 2 as indicated by wound edge swelling and fusion to the other wound side resisting the natural forces of primary skin contraction that tend to widely separate the wound edges apart .The welding was successful only by use of Continuous or Single pulse, spot by spot welding techniques but no human skin wound welding ever have happened by 980 nm diode laser acting in repetitive pulse mode despite peak powers up to 20 watt was adopted with pulse duration from

0.05 sec. up to 5 seconds at pulse repetition time of 50 msec. giving a maximum repetition rate that the device can deliver. i.e. 20 pulse per second and utilizing a wound scanning laser beam movement speed at 4 mm / second.

Ironically ,although the laser welded skin incisions was successfully closed by laser , the wound closure was of very low tensile strength so that those laser welded wounds could be reopened easily upon even a slight traction force applied to one side of those welded wounds (Figure 6, 7,8) ,in other words, the welding was of no clinical significance since it would not be able at all to withstand normal daily activity movement of the live patient .In fact, this weakness didn't allow even an intact biopsy excision of the welded wound in order to examine the weld by electron microscope study to confirm that it did actually happen.

Table (2): 980 nm laser parameters of a successful Skin wound welding

Fluence , Power density	Pulse duration/Sec.	Laser Exposure time/Sec.	Laser mode	Power /watt
0.96 watt/cm ²	-----	360	CW at 4 mm/ scan	1
15.9 watt/ cm ²	-----	240	same	2
39.8 watt/ cm ²	120	same	5
63.7 watt / cm ²	60	same	8
79.6 watt / cm ²	30	same	10
103.5 watt / cm ²	10	same	13
199 Joule / cm ²	5	Single pulse per spot	5
254.7 joule/cm ²	4	same	8
278.6 joule/cm ²	3.5	same	10
254.7 joule/cm ²	2	same	16
159.2 joule/cm ²	1	same	20



Fig. (6): Skin wound edge swelling indicating coagulation of the collagen and a successful though weak wound welding.



Fig. (7): Despite a successful wound welding, it was of low tensile strength.

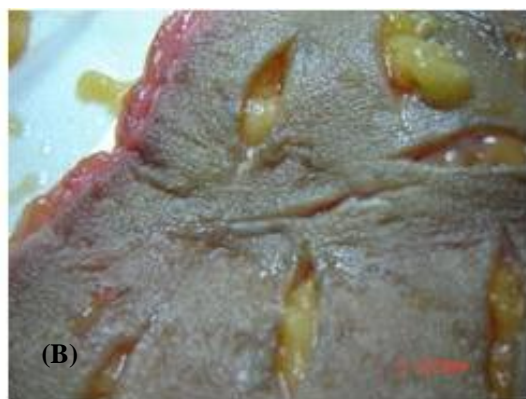
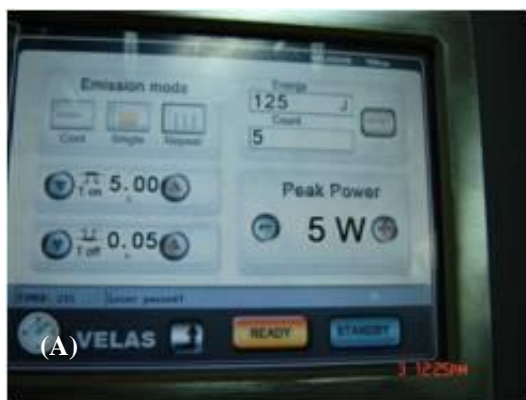


Fig. (8): A successful wound welding yet it could easily be separated by slight traction force applied on one wound side.

Although the utilization of even higher power setting or longer exposure times may results into a stronger welding but this may increases the risk of undesirable thermal wound edge damage too in the absence of real time temperature control feedback systems. In conclusion, they mean that there isn't any particularly unique ideal laser parameter for human skin wound welding but in fact several successful combinations.

The aim should always be to choose the most effective parameter that will yield a successful

strong full thickness transmural wound closure yet it prevent any collateral thermal damage to the wound edges. This can be achieved only by simultaneous real time temperature measurement and /or computerized feedback control systems that will automatically change the laser device power irradiating the target wound.

Discussion

In this study, human skin wound welding was tested with a 980-nm laser system, for the first time. The laser light is converted in the tissue into heat, and this heating is what is believed to cause the structural changes in skin dermal collagen leading to welding.

The justification for carrying out this study on *ex vivo* skin samples rather than on a live human being is to avoid plasma oozing from the skin wound edges that may fill the wound gap and disguise the laser welding process by transferring it into a blood soldering process assisted by laser instead of an actual welding process. Actually that might have happened with many researchers who claimed a strong skin welding by laser in their live animal models despite that they mostly didn't undertake any measures to decrease blood ooze in the wound region in their *in vivo* studies (Murat 2006, Simhon 2007, Zajac 2008).

However, by choosing *in vitro* human skin specimens for this study, the cooling and hydrating effects of the continuous blood flow normally present in the live human skin have also been lost and therefore, the laser parameters applied in this study may not be directly applicable to be used in a live human skin incision closure in future *in vivo* studies. The skin sample used in this study was obtained from one patient only to minimize the effects of individual variations in human skin optical and thermal properties that may influence the photo thermal effects of laser on the skin and hence induce bias in results of this study since In Iraqi population, there are more than 4 Fitzpatrick types of human skin, each of which have its own optical and thermal skin tissue properties. Additionally, human skin thickness and properties vary also with variation of the body region even in the same individual (Charles 2007). Ideally, those *in vitro* skin samples used in this study should have even been slowly heated up to 32-37 C° to mimic the normal living human skin surface temperature since this

will affect the total amount of laser energy needed to raise up the skin sample tissue temperature for welding or soldering to take place but unfortunately, this was technically difficult since adoption of either a dry heating method (e.g., a warm air blower) or a wet heating method (e.g. a water bath) would change the hydration status of the skin itself and hence its optical properties.

Unlike in most other studies (Abergel 1986, Wider1991, A Murat 2006 , Bassam 2009) dental tweezers, rather than sutures were used to temporarily approximate the created skin wound edges while being lasered . This was to avoid adding an extra operative time and to avoid suture trauma or difficulty of placement. Moreover any placed suture material might burn when laser pass over it leading to loss of the suture tensile strength and wound gapping again prior to successful laser welding /soldering has taken place. The curvature of the end of the dental tweezers was found to be even more beneficial than the straight end of surgical tweezers by avoiding the laser path while holding the wound edges together. Other researchers indeed have even started to use specially designed clamps for this purpose (Simhon 2006).

Laser tissue welding is a difficult process. As the process is not well understood, it is difficult to set laser parameters needed for optimum weld strength and to determine when the weld has been achieved. Energy levels and exposure times that may work well with certain tissues may not be the best for other tissues or situations. Hence , it's quite difficult to compare the results achieved by various researchers with this one especially human rather than animal skin samples were used here for the first time.

Moreover, other researchers, have used different wavelength lasers such as 1064 nm or 1320 nm ,1470 nm or 1980 nm claiming that these may even penetrate deeper in the skin (Capon 2001, Özgür 2006, Temel 2009) . Although many other tissue welding studies have used a more water targeting laser like CO₂ laser (McNally 1999) , most of the laser energy in this case would then be absorbed in the outermost layers of the skin tissue because the absorption coefficient of water at 10600 nm is much higher than that at 980 nm, and thereby ,little of that energy penetrates to the deeper skin layers, thus it may result in a dangerous overheating of the superficial epidermal layer while the deep dermal layer ,the target layer for

skin wound welding process, may not heat sufficiently enough to allow the collagen fiber denaturation process to take place, a prerequisite of a successful skin welding process to succeed. Ironically, this relatively poor water absorption of 980 nm laser thus turns to be an advantage by allowing a deeper penetration of this laser into the skin. Additionally, this laser wavelength of 980 nm also has an absorption peak by hemoglobin of blood in future in vivo experiments. This means that even a much higher absorption of laser energy will happen in the vascular dermal layer in the living human skin.

A 4 mm spot size diameter was adopted in this study to ensure an even much deeper penetration of the 980 nm laser into the skin .It is necessary to reduce attenuation of the laser beam caused by light scattering in the subsurface skin layers and, thus, to provide the deepest and most uniform heating of the target site. Moreover, a large spot size also would ensure simultaneous heating of both wound edges to ensure native collagen intertwining necessary for a strong welding process to take place (Coste De 1992 , Nathaniel 2000, Tang 2000, Wolfgang 2002). All other researchers have tried to laser weld wound lengths that are small, like 1-2 cm ,in which the skin primary contraction forces ,due to the skin elastin content ,will not be so strong to gap the wound edges apart .This short wound length was noticed in all researched done in laser assisted wound closure even when the animal model was a huge one like swine , despite the fact that surgical operations ,other than laparoscopic surgeries rarely utilize such short wounds .

That is why, in this study, wounds were made a little longer about 3-4 cm, to ensure that wound edges will separate away enough by primary skin contraction. By doing so, laser skin wound welding has proved to be of low tensile strength and will not be able to withstand the normal daily activity movements in live subjects. Indeed, this seems to be due to small thickness of the collagen containing skin layer, the dermis, in human. This low initial tensile strength of laser welded wounds seems to be an accompaniment of this wound closure method in almost all studies that was done in this regard.

Ironically, in an attempt to overcome this low initial tensile strength of laser welded wounds, some other researchers did place even subcutaneous sutures or even surgical adhesive dressing or tapes over the wound after laser

assisted wound closure, to take away any tension on this welded wound hoping to safely bypass the low initial tensile strength period of laser welded wound (Christopher 2001, Yoko 2005).

It's quite inappropriate to compare the laser parameters utilized to achieve human skin incision laser welding in this study with other animal model based studies since human skin differs a lot from animal skin in layer thickness, blood flow and optical properties. There is paucity in literature for studies conducted on real human skin especially using the same laser wavelength. However, what is quite astonishingly noticeable is that the power densities that have successfully welded wounds in live animal skin in those studies are very close to those that this study has concluded for human skin. Again, this may be misleading due to the absence of the cooling and hydrating effect of blood flow in this *in vitro* study compared to others *in vivo* animal studies (Murat 2006).

Theoretically, single pulse per shot, spot by spot welding using even much longer pulse durations with lower peak powers may result into a stronger wound closure, however, this would be a difficult approach in this study in absence of real time temperature measurement and /or control system since the wound edge skin layers may then be excessively heated, and hence get thermally damaged. The thermal relaxation time of the skin should always be respected to avoid any delay or interference with the post welding wound healing process that would take place.

The most sticking result that this study have revealed is the inability to weld any wound by laser acting in the repetitive pulse mode at the tested laser parameters. Obviously, the repetitive pulsed laser photo thermal effect was indeed very localized to the target site and that there was no enough time for heat diffusion from this target site to the surrounding wound margin tissue to take place that would normally allow the intermingling of the deep dermal layer heat denaturated semi liquid collagen fibers (that actually become a gelatin) with the heated albumin of the solder that upon cooling down again would form a single one strong solid cement like substance that strongly bond the wound edges together. Ironically, the undesirable heat diffusion away from the laser intended target site which is responsible for all undesired collateral thermal damage in most

clinical laser applications become a necessity in laser assisted wound closure process. The way of skin heating obtained by this laser acting in repetitive mode is not synonymous with continuous mode action even when maximum pulse duration and repetition rate was used.

Review of literature showed a paucity of published researches using this mode of laser action in laser assisted tissue welding /soldering. Temel Bilic et al in 2009 study claimed rat wound welding using a continuous and repetitive pulsed mode laser irradiation at 200 ms on 200 ms off pulses, each mode for 5 second exposure from a 1980 nm Thulium laser.

The results also showed weaker wound closure though less tissue damage signs in repetitive pulsed mode than in continuous mode even at same power setting. That study also revealed that wound welding using this 1980 nm laser can be achieved at very low power of 100 mW only 34.6 W/cm^2 (Temel 2009).

To sum up, in order to achieve a strong wound welding then laser acting in continuous mode at some ideal parameters would guarantee this to happen but ironically, at same time, it may cause thermal damage and delays wound healing process in absence of a suitable real time thermal measurement feedback laser power output control systems. A low powered setting with long exposure time is thus recommended. The majority of previous skin welding studies have used either continuous-wave (CW) delivery of radiation or temperature-controlled welding systems with constant surface temperature feedback. Use of laser in pulsed mode in spot by spot welding, on the other hand, though would be much safer and friendly for the wound margins but would not result in a wound closure neither as strong as nor as water tightly sealed as that obtained by continuous mode lasers. Even in this approach, a relatively long pulse durations are required and thus it appears as if laser is acting in an interrupted continuous mode heating fashion rather than a homogenous heating of the entire dermal skin layer or the solder as when it takes place in an actual continuous mode laser irradiation.

Fried et al. actually also have concluded that continuous laser irradiation actually yields stronger wound closure than spot by spot laser irradiation method. They have applied a strong absorber (India ink) to the apposing edges of the incision and scanned the laser beam rapidly across the weld (Nathaniel 2000). The scanned CW laser welding system thus has provided

several advantages over a spot welding technique, including a smoothing out of inhomogeneities in the laser beam profile and a more uniform delivery of radiation along the weld site.

Human skin incision welding didn't happen using the same laser parameters that claimed to be successful for welding the rat wounds in Murat et al study, i.e., 6 W peak power and 0.4 sec. pulse duration per shot utilizing the spot welding technique (A Murat 2006). In addition to the differences between human skin and rat skin in structure and optical properties, A Murat et al experiment was done in vivo in which there was a continuous blood supply cooling the area.

Eventually, the blood may have filled the wound gap and lead to laser assisted blood coagulation thereby joining the wound edges together, rather than an actual laser skin welding process as our rat skin welding experiment did show. His main aim was to study the healing pattern in laser welded wounds.

In fact, most studies that have been carried out on live animals aim to study either the healing pattern or the tensile strength of the laser welded/soldered wound. A quick review of them revealed an initial delay phase that happens in the first few days following the laser wound welding in comparison to standard suture closed wounds indicating that there was always some laser induced thermal damage or at least inflammation of the skin margins that delays the start of the wound healing process .

This then have lead to a temporary decrease in the wound tensile strength in first few days following the laser action on the wound .Several researchers have claimed that this delay has been avoided when real time temperature measurement control systems were adopted in laser assisted wound closure (Brosh 2004, Christopher 2001, Lauto 2001, Nathaniel 2000, Simhon 2004).

The most striking advantage of laser assisted wound closure that this study has confirmed so far is that it could really be 4 times faster than conventional technique of skin wound closure just like other researchers has found (Christopher 2001).

Conclusion

A 980-nm diode laser wound welding proved to be of low tensile strength to be useful in clinical practice though it can really dramatically shorten the operative time required

to close a wound if proper laser parameters were used among an endless effective combinations of laser parameters.

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لحام جرح جلد الإنسان بمساعدة ليزر أشباه الموصلات 980 نانومتر: دراسة تجريبية خارج الجسم الحي

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الخلاصة
ان لحام الجروح بمساعدة نو مزايا وفوائد عديدة مقارنة بالطرق التقليدية. أن الغرض من هذه الدراسة خارج الجسم الحي التي أجريت في معهد الليزر للدراسات العليا في جامعة بغداد هو تحديد مدى فاعلية ليزر أشباه الموصلات 980 نانومتر في المساعدة في لحام جروح الجلد لدى الإنسان . لقد استخدم في هذه الدراسة عينات من جلد جدار البطن المستأصل من عملية تقويم جدار البطن لمریضة تبلغ 45 سنة من العمر. تم استحداث عدة جروح طولية قطعية بطول 3-4 سم كاملة العمق خلال عينة الجلد هذه ثم تمت محاولة لحامها بتعريضها لليزر بطول موجي 980 نانو متر وقطر البقعة 4 ملم باستخدام طاقات وأنماط عمل مختلفة . لقد حصل لحام لجروح الجلد ولكنه ذو قوة ضعيفة عند استخدام الليزر عند طاقات وأنماط عمل مختلفة. لقد أظهرت النتائج انه رغم نجاح حصول لحام هذه الجروح باستخدام طاقات وأنماط إشعاع ليزر مختلفة إلا ان هذا اللحام لم يكن بقوة كافية ليكون ذا فائدة سريرية للمرضى.