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Laser Welding of Enterotomies in Eversion

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Abstract

Twenty-one albino rats (average weight of 152 grams) were studied by inducing 28 full thickness enterotomies (involving from half to full circumference of the rat small intestine). Trials at welding of the enterotomies using Nd-YAG laser non-contact fiber were done using 5 watts power and duration ranging from 5-8 seconds as a continuous wave in spot applications. Simple nontoothed forceps was used for tight approximation of the bowel ends in eversion. A new visual end point for lasing was noticed during welding. Results were presented and analysed.

Introduction

FOR a long time the sutures and/or staples were the only means available to the surgeon for constructing intestinal anastomoses **[1]**.

The need for an alternative technology for tissue approximation is becoming more apparent in the last few years, specially after the wide adoption of laparoscopic surgery, where suturing requires a considerable practice on a trainer **[1]**.

The use of low-power laser energy to fuse gastrointestinal tissue had recently been shown to provide an alternative to conventional suture or stapling techniques **[2,3]**.

In those studies, welding was done in inversion using tissue blanching and shrinkage as a visual end point for lasing resulting in a leakage rate of 14.28% due to thermal damage at the site of welding **[4]**.

In this study, we tried to assess the ability of laser power alone to weld enterotomies of various dimensions in eversion using simple nontoothed forceps for tight tissue approximation.

Material and Methods

Twenty-one albino rats with average body weight of 152 grams were included in this study. Anaesthesia was induced by injecting each animal with 0.2-0.3 ml chloral hydrate intraperitoneally.

Using standard operative techniques and a midline incision, the small intestines of the animals were pulled out. Twenty-eight transverse enterotomies were made by scissor cuts. Twenty cuts involved 1/2 of the circumference, 5 involved 3/4 of the circumference and three involved the whole circumference of the rat small bowel.

A neodymium-yttrium aluminium garent (Nd-YAG) (1.06 μm), laser system (Heraeus laser sonics, model 6000) was used to weld the enterotomies, using 5 watt power over 5-8 seconds period. The laser beam was delivered via a 400 μm fiber at 1 cm distance from the enterotomies making a spot of 0.5 mm and an energy density (ED) of 31.826-50.92 joules/cm^2. Tight tissue apposition of the edges of the enterotomies was done in eversion using a nontoothed forceps on each side of the welding area without the use of stitches, staples or intraluminal stents during laser application. A continuous drip of normal saline at room temperature (1 drop/second) was run on the tissues for cooling during fusion. Energy was delivered as a continuous wave in spot applications (3-4 spot applications were used in order to weld each 1/2 circumference of the rat intestine.

A silk marker stitch 2/0 was passed in an avascular area of the mesentery and tied loosely at the site of welding in order to facilitate identification of the welded site later on.

Success or failure of the welding trials were judged intraperitoneally by the naked eye. The end point of successful welding was complete sealing of the enterotomy by melting of the mucosa on each side to fill the gap.

Animals were sacrificed at days 0 (the day of the operation), 3 and 14 postoperatively. The anastomoses were inspected by the naked eye at the planned time of sacrifice for leakage, abscess formation, inflammation, adhesions, luminal stenosis or for proper healing. The welded enterotomies were excised and fixed in formalin for later paraffin embedding and sectioning . Sections were made perpendicular to the lased surface. Sections of 5 μm thickness were studied histologically after staining by haematoxylin and eosin stain, Mallory and Masson trichrome stains [5,6].

Results

Four visual evidenced patterns of success or failure of the welding trials could be detected intraoperatively (Table 1).

The five enterotomies in pattern "3" were sutured intraoperatively after failure of sealing and separation, then 1/2 circumference enterotomies were done at another site in the small bowel of the same animals and welded with laser.

The four animals in pattern "4" died in the postoperative period prematurely as a result of anastomotic leaks and peritonitis (2 at day 1 and 2 at day 2).

Microscopic examination of the laser welded enterotomies sacrificed on day 0 (the day of the operation) showed the formation of a plug formed of necrotic material and collagen filling the gap between the two everted edges (Fig. 2).

On day 3, the laser site revealed minimal cellular infiltration with granulation tissue formation. The surface mucosa began to regain its continuity with some collagen fibres bridging the anastomotic gap (Figs. 4, 5). The goblet cells lining the regenerated mucosa were numerous when compared with those lining the enterotomy

Table (1): The Four Visual Evidenced Patterns of Success or Failure of the Welding Trials Detected Intraoperatively .

	1/2 circumference enterotomies (n = 20)	3/4 circumference enterotomies (n = 5)	Complete enterotomies (n = 3)
1. Complete sealing* of the gap	14	3	-
2. Inadequate sealing without separation	2	-	-
3. Inadequate sealing without separation	-	2	3
4. Thermal damage (blanching despite complete sealing)	4	-	-

* Complete sealing means achieving the end point of welding, i.e. melting of the mucosa from both ends to fill the gap without tissue blanching or shrinkage (Fig. 1).

at day 0 (Figs. 3, 5). residual collagen was deposited between the regenerated villi and crypts.

Fig. (1): A photomicrograph of a formalin fixed specimen of the small intestine of a rat sacrificed at day 0 (day of the operation)

Fig. (2): A micrograph of a laser welded enterotomy in the small bowel of a rat sacrificed at day 0 showing a plug formed of collagen (C) and necrotic material (n) filling the gap bwtween the two everted edges (Masson trichrome, original magnification x 40).

Fig. (3): A photomicrograph of a laser welded normal distribution of goblet cells in the epithelial lining of the everted edges of the gap (Hx. & E., original magnificantion x 100).

Fig. (4): A micrograph of a laser welded enterotomy in the small intestine of a rat sacrificed at day 3 postoperative showing regenerated intestinal villi (V) and crypts (Cr) filling the gap. The lining epithelium showed numerous goblet cells. Residual collagen (C) is present between the regenerated villi and crypts. The submucosa shows deposition of collagenous fibres (Masson trichrome, original magnification x 100).

By day 14, complete sealing of the gap with the regenerated small intestinal mucosa (inside) and collagen fibres (outside) was noticed with the deposition of collagen between the villi and crypts (Fig. 6a & b). Both the regenerated short intestinal villi and the regenerated numerous crypts were lined with numerous goblet cells. Minimal mononuclear cell infiltration was also detected in healed area (Fig. 7).

Fig. (5): A photomicrograph of a laser welded anatomosis at day 3 showing minimal cellular infiltration and granulation tissue formation (arrow). Collagen (C) is deposited between the regenerated small intestinal villi and crypts. Numerous goblet cells line the regenerated mucosa (Hx. & E., original magnification x 100).

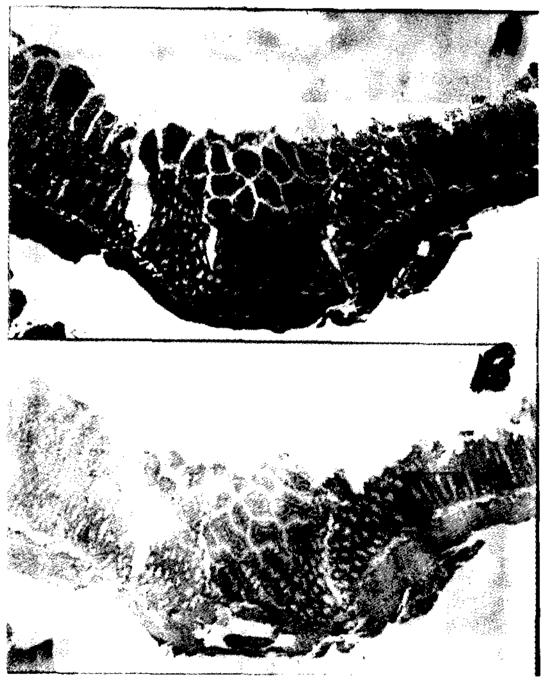

Fig. (6) Photomicrographs of laser welded everted anastomosis of a rat sacrificed at day 14 showing that the anastomotic defect is almost completely healed by the regenerated collagenous fibres (C) and the regenerated mucosa. The regenerated crypts (Cr) appeared numberous. Residual collagen (C) was deposited between the short regenerated villi (V) and the numerous crypts (Cr) (A: Mallory trichrome, original magnification x 60, B: Hx. & E., original magnification x 40).

Fig. (7): A micrograph of a laser welded enterotomy of the small intestine of an albino rat sacrificed at day 14 showing numerous goblet cells lining the regenerated mucosa and the deposition of residual collagen (C) between the regenerated villi (V) and crypts (Cr). Minimal cellular infilration was noted in the healed area (Hx. & E., original magnification x 100).

Discussion

The use of low power laser energy to fuse gastrointestinal tissue had recently been shown to provide an alternative to conventional suture or stapling technique [3,4].

The mechanism of laser tissue fusion has not yet been elucidated. Electron microscopy has provided evidence for possible crosslinking of collagen [7].

Good control of laser fluence and good tissue apposition are essential prerequisites for welding enterotomies [8].

Although many authors recorded that good tissue apposition could be achieved by sutures [3] (laser assisted intestinal anastomosis) or using biodegradable intraluminal stents [2], in order to achieve a tight serosa to serosa coaptation for sutureless end to end bowel anastomosis in inversion. They depended in these experiments on a visual end point of lasing evidenced by tissue blanching and shrinkage [3,4]. In this study tight tissue approximation in eversion during lasing using a simple nontoothed forceps was successful in most of the enterotomies involving 1/2 and 3/4 of the circumference of the bowel, but failed to achieve adequate coaptation in enterotomies involving the whole circumference of the bowel. Laser welding of enterotomies in eversion relied upon a new visual evidenced end point which was melting of the mucosa on both sides to achieve complete sealing of the gap (successful welding in 14 out of 14 observed trials "pattern 1", i.e. 100%). When evidence of thermal damage was awaited, i.e. tissue blanching and shrinkage, the animals died postoperatively from anastomotic leaks and peritonitis (pattern "4", n = 4).

Histological studies of our laser welded enterotomies in all days of sacrification (0, 3 and 14) revealed the involvement of collagen in the mechanism of bridging and fusion of the gaps. This result agreed with that of other investigators [4,9] who suggested that VI collagen played a role in the fusion process when they observed a homogenizing change in periodically banded collagen with interdigitation of altered individual fibres that appeared to be the structural basis of the welding effect in microvessels fused with 1,319 nm Nd-YAG laser. They suggested that type VI collagen which coats the outside of the periodically banded type I fibres may be involved in the interdigitation of those fibres.

In some previous experiments [4,10], comparison between healing of sutured enterotomies and healing of laser welded ones revealed the presence of intense inflammatory process, adhesions and increased granulation tissue formation in sutured wounds due to foreign body reaction caused by the sutures. They proposed that an advantage of laser fusion over conventional repair is the lack of foreign body reaction in the laser welded enterotomies. The minimal cellular infiltration and absence of granuloma formation observed in sections examined from the sutureless laser welded anastomoses at days 3 and 14 in the present study confirmed the above mentioned findings.

Microscopic examination of the regenerated mucosa filling the gap of the enterotomies in animals sacrificed at days 3 and 14 showed numerous regenerated crypts that might be due to the ability of the crypts to produce a continuous supply of new cells which progress up the villi where they mature before degenerating and being shed [11].

In the present study, an increase in the number of goblet cells lining the regenerating intestinal villi and crypts shown in sections obtained from enterotomies at days 3 and 14 postoperatively might be due to a protective mechanism of the regenerated mucosa against tissue trauma after the process of cutting and welding.

We conclude that welding of the bowel ends using laser in eversion will provide a new, relatively safe visual end point which is more acceptable than the visual end point of welding in inversion which signifies the start of thermal damage.

The use of simple forceps for tissue approximation during welding of enterotomies in eversion would simplify the technique for its possible use in the future through the laparoscope.

However, further work is needed in order to overcome the inability of non-toothed forceps to approximate tissues over a long area.

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