IIOAB JOURNAL
ISSN: 0976-3104

ARTICLE

EFFECT OF LOW LEVEL (INFRA RED) LASER THERAPY ON THE SURGICAL WOUND HEALING IN HAMSTER

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ABSTRACT

Back ground and objective:*Accelerating wound healing is a very important factor for patients to return to ordinary life. Due to previous studies, Laser seems to have positive modality effect on cutaneous wound healing. This study aims to investigate the effect of infrared light laser on cutaneous surgical wound in hamster and to compare factors such as angiogenesis, number of fibroblasts and collagen formation with control group* ***Materials and methods:*** *This was an experimental study which was carried out with correlation of Quazvin University of medical science with MEHREGAN pet clinic in winter of 2009. Twenty -five suri hamster were randomly allocated in two groups and four parallel wounds was made on their back, Case group was radiated by red-light laser 630 nm. After 2/5/10 and 14 days, skin biopsy were obtained and number of vessels , fibroblast and collagen production were compared with control group.* ***Result:*** *Evaluation of wound in experimental and control groups showed significantly increased number of vesseles and fibroblast as well as collagen production in laser radiating group.* ***Conclusion:*** *Our study suggest that laser therapy can accelerate wound healing in compression with control group.*

KEY WORDS
Laser,Wound healing, Angiogenesis, Fibroblasts, Collagen formation.

Published: 15 October 2016

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INTRODUCTION

Acceleration of cutaneous wound healing is a very important factor after surgery for surgeons for returning to ordinary life style in earlier time. Wound healing is complex phenomenon involving a number of processing including induction of acute inflammatory process, migration and proliferation of parenchyma and also connective tissue cells, synthesis of ECM proteins, remodeling of connective tissue and parenchymal component and then colagenization and acquisition of wound strength. [1,2,3] After day 3, granulation tissue progressively invade the inicision space and collagen fibers start to present. By day 5, neovascularization is maximal and fibroblasts proliferate. During the second week , accumulation of collagen and proliferation of fibroblasts is continued. [4]
Low level lasser therapy is a form of phototherapy that invades the application of low power monochromatic and coherent light to lesions to stimulating wound healing. It can increase the speed, quality and tensile strength of tissue repair [5]. The effect of LLLT are photochemical, not thermal and the response of cells due to changes in photo acceptor molecules (knows as chromophores) [6].

The exact mechanism of action of LLLT has not been completely understood but it can stimulate metabolism [7]. The effect of LLLT including wound epitelialization, reduction of edema and inflammation, increase granulation tissue, increase fibroblastic prolferation, increase exteracellular matrix synthesis and neovascularization, all of which lead to better tissue oxygenation and nutrition and enhance wound healing [8,9,10] Previouse studies revealed that the amount of total collagen was significantly increased in leser treated wounds over control group also it accelerates the production of collagen. [11]

In this study we evaluate the effect of infra red-light laser nm in wound heading in hamster and compare with control group
.

MATERIALS AND METHODS

This was an experimental study which was carried out in small animal clinic of Mehregan and Qazvin University. All animal procedures were in accordance with the declaration of Helsinki and the guide for the care and use of laboratory animals.

We performed our study on a Total 25 suri hamster (random male and female). They were randomly divided into two groups including 15 experimental and 10 control groups . After anesthesia we perform four parallel surgical scrap on the back of hamster measuring each one 1cm in both groups . Red light laser nm was radiated 6 times, each time 10 seconds to 15 hamster of experimental group .
Then biopsy were taken in day second, days fifth, tenth and forteenth after radiation from both experimental and control groups.
Sample were sent in formalin 10% to laboratory. Tissue processing and sectioning of paraffin embedded tissue were done and then slides stained with H&E (hematoxylin and Eosin).

Sides were studied by pathologist. Number of vesseles in field 40 was counted in day second and day fifth samples in both groups.
Also median number of fibroblasts in field 100 was counted in day fifth and day tenth samples in both groups. For day forteen specimen samples , Trichrom staining were performed and qualitative estimation for collagen formation in two groups were done .Data were analyzed through independent student T. test using SPSS version 10 and statistical P value less than ./.5% was considered significant and in non normal distribution . Kolmogrov- Smirnov test performd for data such as number of fibroblasts in day fifth and tenth.

RESULTS

Histologic evaluation of wounds in two groups and the statistical analysis of data revealed significant difference between control and laser treated hamsters in mean number of vesseles and number of fibroblasts [Table 1] .

Table1: number of vesseles in control and laser treated group in day second samples

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Control	18	18	20	18	19	20	19	21	18	22	19	17	20	13	25
Laser radiated	23	23	27	23	26	26	25	28	24	29	23	22	27	22	30

Table 2: Mean and SD of vessele number in two groups in day second samples

SD	Mean of vesseles number	Group
2/03	19/47	Control
2/6	25/2	Laser treated

(P=./001%) in independent sample T test

Second day samples show mean number of 19/4 of vesseles in control group in comparison to laser treated group with mean number of vesseles of 25/2. T test show significant difference between two groups (P<%015) [Table 2].

Table 3: Number of vesseles in day fifth

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Control	35	32	30	35	31	29	33	31	30	34	32	30	29	35	34
Laser radiated	40	36	30	40	34	30	38	35	32	40	40	35	30	40	40

Table 4: Mean and SD of vesseles number in two groups in day fifth

SD	Mean of vesseles number	Group
2/2	32	Control
4/05	36	Laser treated

In fifth day samples also mean number of vesseles in 15 hamster was 36 in red light radiated group which show significant difference in comparison to control group . [Table 3,4] and [Fig. 1]

Fig. 1: It shows laser radiation of wound in hamster.

Table 5: Number of fibroblasts in day fifth

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Control	60	65	60	60	61	63	61	59	62	60	64	61	60	61	62
Laser radictor	62	74	62	65	67	70	68	60	70	60	71	70	65	65	70

Table6: Mean and SD of fibroblasts number in day fifth

SD	Mean of fibroblast number	Group
1/67	61/27	Control
4/31	66/6	Lased treated
4/31	66/6	Lased treated

P<0/001 in Mann- Whitney test

Number of fibroblasts estimated in experimental and control group (61/2 and 66/6) which show significant deference in Kolmogrov – Smirnov test between two groups.(P<0/0001) [Table 6] and [Fig. 2]

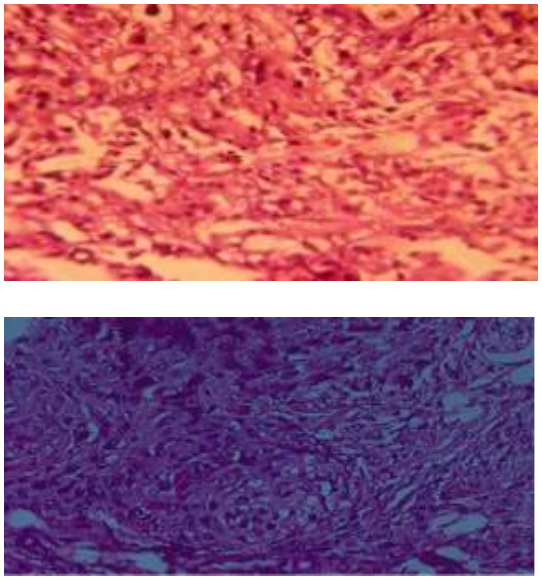

Fig. 2: Comparision of vessels number in fifh day in control and laser treated group.

Table 7: Mean number of fibroblasts in day tenth samples

SD	Mean of fibroblasts number	Group
1/64	53/37	Control
2/12	71/73	Lased treated

P<0/001 in Mann- Whitney test

In day tenth samples, mean number of fibroblasts in two groups show also significant difference with the same test (P<0/001)[Table 7] ,[Table 8] Qualitative collagen production with trichrom staining in forteen day samples show significantly increased collagen production in experimental groups in comparison to case group [Fig. 3],[Fig.4]

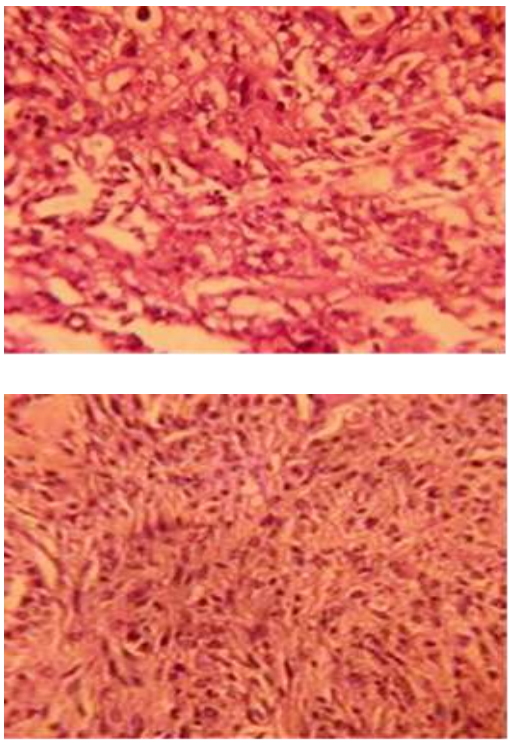

Fig. 3: Comparision of fibroblasts number in day fifth in control and laser treated group.

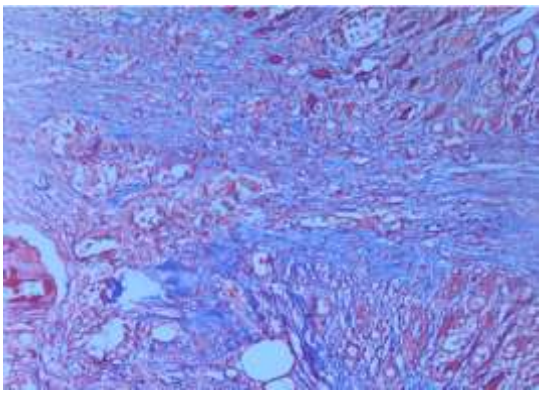

Fig. 4 : Increased collagen production in day fifteenth specimens in laser treated group (Trichrom staining).

DISCUSION AND CONCLUSION

Studies has showed that laser therapy in wounds can increase cellular content and enhance granulation tissueformation ,collagen deposition and fibroblasts production. [12,13]. it is unclear how LLLT works. LLLT may reduce pain related to inflammation by lowering, in a dose-dependent manner, levels of prostaglandin E2, prostaglandin-endoperoxide synthase 2, interleukin 1-beta, tumor necrosis factor-alpha, the cellular influx of neutrophil granulocytes, oxidative stress, edema, and bleeding. The appropriate dose appears to be between 0.3 and 19 joules per square centimetre.[14] Another mechanism may be related to stimulation of mitochondrion to increase the production of adenosine triphosphate resulting in an increase in reactive oxygen species, which influences redox signalling, affecting intracellular homeostasis or the proliferation of cells [15] The final enzyme in the production of ATP by the mitochondria, cytochrome c oxidase, does appear to accept energy from laser-level lights, making it a possible candidate for mediating the properties of laser therapyHelium Neon (632/8 nm)and laser diodes between (633-670 nm) are absorbed by the mithochondria cytochroms (mainly cytochrom C oxidase) and known to act mainly on superficial epithelial tissue due to limited penetration of red light. [16] studies have shown that endothelial cells showed higher cell counts when irradiated with (660 nm)wave length (in coparision with 820nm)It induce fibroblasts to secret growth factors and cytockins,possibly increasing rate of mitosis and reducing cell dead.then they are effective in wound healing. Lasser therapy is a simple and non invasive therapy which can promote wound healing, relef pain and have no reported side effects [17] Accelerating of surgical wound healing help patients recover to normal life . in this study we evaluated the effect of low-level laser therapy (630nm) in a accelerating wound healing in hamster. Our study showed that laser therapy can significantly increase number of vesseles in second and fifth days .Laser can stimulate proliferation of fibroblasts and increase its number (in day 5 and 10). It is significantly effective in collagen production.and can promot process of collagenization (in day 14) .Then our study suggest low level laser therapy can accelerate wound healing in rats.

Our study suggest that low level laser therapy (630nm) can accelerate and promote surgical wound healing in hamsters.

CONFLICT OF INTEREST
There is no conflict of interest.

ACKNOWLEDGEMENTS
None

FINANCIAL DISCLOSURE
None

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