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Original article

Impact of laser irradiation on regenerative processes of full thickness skin

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Abstract

A total of (twenty four adult New Zealand male rabbits), were used in this study, the animals were divided into (two groups); control and treated, (twelve rabbits each). All animals were underwent a whole thickness skin graft transplantation from the lateral aspect of one thigh to the lateral aspect of the other thigh. Treated group was irradiated with Ga.Al.As. diode Laser (904 nm) wave length and power (5mW) for (5min./session), (1cm) distance from the suturing line which was (2x2 cm), dimensions. Irradiation began at the time of transplantation directly and continued for (7 days) after that. Grafts of both control and treated group were examined clinically daily while (four animals) of each group were anaesthetized at the (days 3,7&10) post the operation and the samples collected from them were sent for histopatholgical examination. Clinically all the grafts seen vital and pinkish, while the line of the incision began to disappear after (3 days) of irradiation with diode laser comparing with the control group in which the line of incision began to disappear (7 days) after the transplantation, and the hair began to appear after (7 days) of irradiation with the diode laser while it began to appear (10 days) after the transplantation in the control group. Histopathologically samples manifested little inflammatory cells and proliferation of primitive microvascularization, after (3 days) of irradiation while the inflammatory cells were widly diffused at the site of transplantation in the control group for the dame period, results of examination of the sample collected after (7 & 10 days) revealed more maturation of the blood vessels and hair follicles which were numerous in addition to accomplishment of the epithelial migration while the blood vessels and hair follicles were few and the epithelial migration was rare in the samples collected from the control group for the same period.

Key words: Laser, Skin healing, Skin graft.

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Introduction

Low power laser irradiation has been reported to reduce inflammatory reactions, produce analgesia and promote regeneration and healing. Its mechanism of action is unknown, one hypothesis is that the light produces free radicals which have beneficial effects at low concentrations, (1), they accelerate the collagen and its precursor's maturation's, promote the improvement of microcirculation, (2), they also activate the mast cells, (3). One of these lasers are the diodes, they have been used for therapeutic applications over the last twenty years, and their use expanded numerously due to the better understanding of laser-tissue interaction and the development of sophisticated delivery systems, (4). Experimental works found that the Ga.Al.As. Diode lasers stimulate the human embryonic fibroblast

cells and maximum increase in collagen production and cell biostimulation after (4 episodes) of laser treatment at (24 hours) intervals, (5) while the photobiostimulation effect of Ga.Al.As. Diode laser on the skin grafting suggested much more numerous microvascularization, which is an important indication for the success of the grafting and the early healing of the grafts, (6). In addition to that; grafted skin sometimes develop an aberrant pigmentation which represents an unwelcome complication to an otherwise successful split skin graft resulting in a loss of color match and so it follows cosmetics but laser therapy succeeded in treatment of such a case, (7). Due to the importance of skin grafting and the absence of any local previous study on the effect of Low Level Laser Therapy (L.L.L.T.) on the healing of the grafts, this study was designed to be a preliminary experiment to highlight the effect of Ga.Al.As. Diode laser on skin grafting.

Materials and Methods

A total of (24 adult New Zealand rabbits) were in this study, they were classified in to two groups; control and treated, (12 rabbits each). The animals prepared for the surgical procedure by injecting them with a pre-medication drug; Acepromazine maleate[1] (10mg/kg. B.W.) injected i/m, after (10 minutes) a mixture of ketamine hydrochloride[2] (10mg/kg. B.W.) and xylazine[3](5mg/kg. B.W.) Injected i/m to the animals, (8). The operation began by removing of a whole thickness skin grafts (wolf's graft) of (2x2 cm) dimensions from the lateral aspect of one thigh and placed in a normal saline solution and similar graft from the other thigh was removed and then the graft of the first thigh was transmitted to replace the graft of the other thigh. The grafts fixed to the recipient site with stitches of simple interrupted sutures using surgical silk[4]3-0, the same thing was done on the other side then the animals injected with systemic antibiotics; penicillin (1000 iu/kg. B.W.) and streptomycin (10mg/kg. B.W.) i/m for (3 days) after the operation. Sutures of the animals of the irradiated group were removed (4 days) after the operation while those of the control group removed after (7 days). Animals of the treated group were irradiated using a Ga.Al.As. diode laser[5] with a wave length (904 nm) and power (5 mW) for (5 minutes/session). Sessions began after the operation directly and continue for one week after that, irradiation with the laser done by directing the beam (1cm) distance from the suturing line all around the graft. All the animals examined clinically daily looking for the vitality, elasticity and the color of the grafts while (4 animals) of each group were anaesthetized and skin samples from edges of the grafts were harvested for histopathological study at the ($3^{rd.}$, $7^{th.}$ & $10^{th.}$ postoperative days)

Results

Clinical examination of the grafts of all the animals revealed that they were vital, elastic and pink along the period of the study. The clinical and histopathological differences between the control and treated group are shown in table No.1 & No.2.

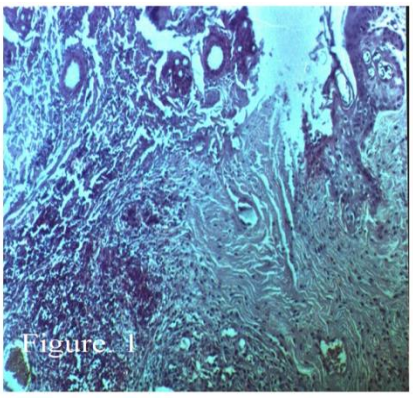
[1]Calmivet, vetequinal, S.A., 7200, Lure (France).
[2]Ketallar, 50mg/ml, Parke Devis co. Gwent, U.K.
[3]*Rompun, 20mg/ml, Partex Holland, B.V.De Hoeve 28*
[4] *Ethicon, Ltd, PO Box 408, Scotland, U.K.*
[5]*Russian – Polish joint Venture, 103030, Moscow, Russia.*

Table .1: showing the differences in clinical findings between the two groups.

Day	*Control group*	*Treated group*
3rd. Day post operation.	The grafts were vital, light pink in color.	The pink color was stronger. The line of incision began to disappear.
7th. Day post operation.	Hair follicles not appeared. The line of incision began to disappear. Absence of the hair follicles.	Hair follicles began to appear with complete healing of the marginal incision.
10th. Day post operation.	Hair follicles very rare with complete healing of the marginal incision.	Hair follicles numerous and the marginal wounds looked finer.

Table. 2: showing the differences in histopathological findings between the two groups.

Day	Control group	Treated group
3rd. day post operation.	Widely diffused inflammatory cells but no evidence of granulation tissue.	Little diffused inflammatory cells. Proliferation of the primitive blood vessels was evident through, with newly formed capillaries were seen. Hair follicles appeared. Migrating newly formed stratified sequamous epithelium could be seen on both sides of the incision, (Fig. 1)
7th. day post operation	Collagen fibers in the dermis were scantly appeared in addition to little newly developed blood vessels while the area still filled with inflammatory cells.	Collagen fibers was abundant and new vascularity was numerous (Fig.2) in addition to hyperplastic stratified sequamous epithelium (Fig.3&4), more hair follicles could be seen, some of them atrophied while others hyperplastic on their epithelial lining, still hemorrhagic spots present in addition to filtering of clots in matrix of the dermis (Fig.5).
10th. day post operation	Few numbers of hair follicles and blood vessels appeared and the migration of the epithelium was still rare and seen as separated patches.	Numerous well-developed hair follicles and capillaries appeared, epithelial creeping complete and hyaline connective tissue was abundant.



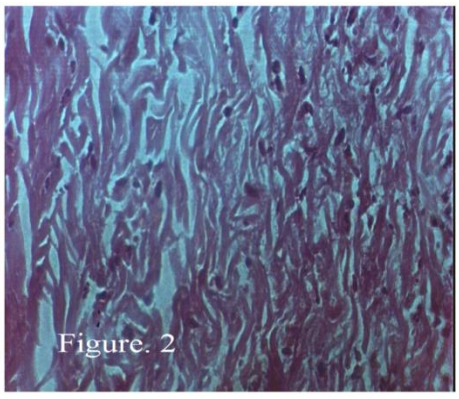


Figure.1: Treated group (3rd. postoperative day), appearance of the hair follicles beside the original skin area (H&E X 20*).

Figure.2: Treated group (7th. post operative day) abundance of collagen fibers at the dermis with numerous new vascularity, (H&E X 40).

*Original magnification

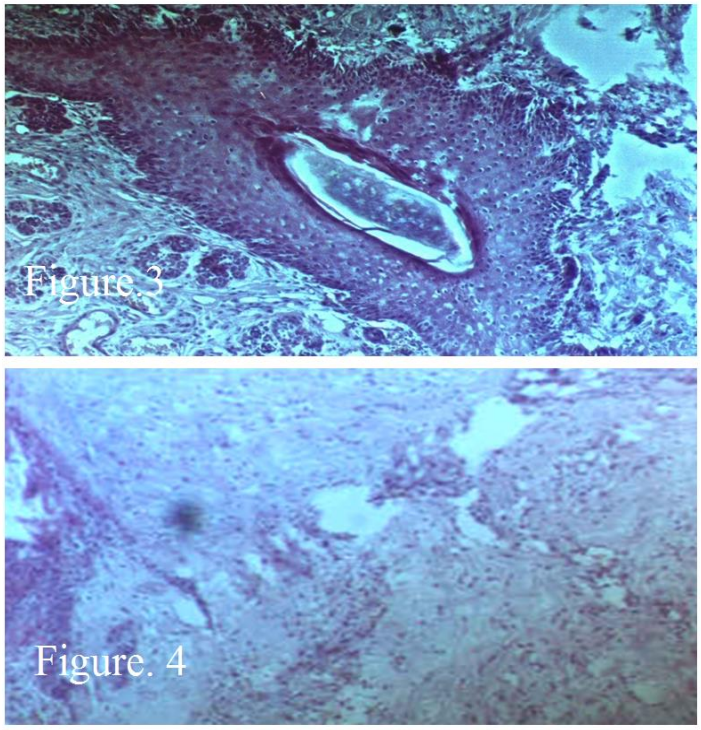


Figure.3: Treated group (7th. post operative day) relatively thick matrix with good differentiated and developed epidermal layer at the line of the incision, (H&E X 10).

Figure.4: Treated group (7th. post operative day) hyperplasia of the epidermis, note its normal cellularity, (H&E X 40).

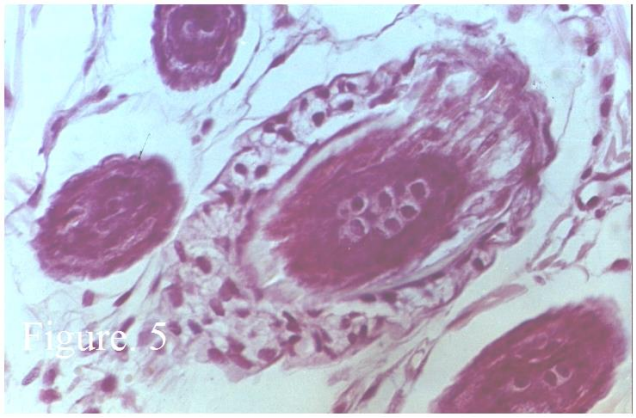


Figure.5: Treated group (7th. post operative day) hyperplasia of the hair follicles deeper in the dermis, (H&E X 40).

Comments

For a skin graft to survive successfully it needs a rapid establishment of a new circulation, (6 & 9), so the most important note should he mentioned for any skin graft transplantation whether it is carried out is vivo or in vitro is how to prepare a good microvascularization in the bed of the graft immediately after placing the donor graft in the recipient site. Results obtained from the treated group after (3 days) of irradiation revealed that regenerative processes began in this group earlier compared with the control group, these findings can be attributed to the stimulating effect of the laser therapy

which enhances the phagocytic activity of the leukocytes in the blood and creates anti-inflammatory effects, the phagocytes help in getting rid of the dead tissues and prepare the site for new tissue formation, this fact explain why the site of the operation was widely diffused with inflammatory cells in the control group while just little inflammatory cells could be seen at the site of the operation in the treated group,(10). The early proliferation of the primitive blood vessels and well developed capillaries as well as the migration of newly formed stratified sequamous epithilieum and appearance of hair follicles in the treated group, laser therapy significantly enhances the microvascularization of the graft in the irradiated rats using flourescin angiograply, (11). The line of the incision began to disappear after (3 days) of irradiation while the healing of the marginal incision accomplished after (7 days) of irradiation, these finding can be explained by the increase of the portion contents of oxygen in the tissues and by stimulation of the mitotic cell activity as well as the bactericidal effects which are exerted by the action of the laser therapy, these findings are in agreement with those obtained by, (12). Samples obtained from the treated group after (10 days) showed that the site of the operation retain its normal cellularity, these findings supported by (13) who mentioned that laser therapy accelerates the healing of perforated wounds, i.e. those involving whole skin thickness, (13), so if the graft margin of the present study considered as a wound of skin involving all the layers, the healing acceleration ability of this wound in the treated group can be understood by increase collagen formation, re-epithilization and mast cells activation which promoted by the laser therapy, activation of the mast cells help in release of mediators which may be one of the mechanisms by which the L.L.L.T. accelerates the tissue repair, (3).

Bibliography

1. Iwase – T; Hori – N; Morioka – T and Carpenter, Do., (1996). Low power laser irradiation reduces ischemic damage in hyppocampal. Laser - Surg. - Med., 1996; 19 (4), pp. 465-70.
2. Ihsan, F.R.Mohammed. Low-Level Laser Therapy Accelerates Collateral Circulationand Enhances Microcirculation. Photomedicine and Laser Surgery: Vol. (23), Issue (3) , June 14 2005. Pages: 289–294.
3. Al-Sayed – SO. & Dyson, M., (1996). Effect of laser pulse repetition rate and pulse duration on mast cell number and de-granulation. Laser – Surg – medical, 1996, 19 (4): pp.433-7.
4. Milroy – BC., (1997). Laser and Skin diseases, what works and what does not. Aust-Fam-physician, 1997; sept; 26 (9), pp. 1037-40.
5. Skinner – SM; Gage – JP; wilce – PA and shaw – RM., (1996). A preliminary study of the effect of laser radiation on collagen metabolism in cell culture., Aust – Dent – J., 1996, Jan; 4 (3): pp. 188-92.
6. Ihsan, F.R. Mohammed, Nuha, Al- Mustawfi & Laith, N.K.; Promotion of regenerative processes in injured peripheral nerve induced by low-level laser therapy. Photomedicine and Laser Surgery: Vol. (25), Issue (2) / 2007. Pages 107 – 111.
7. Raine – C; al-Nakib – K and Quaba – AA., (1997). A role of lasers in the treatment of pigmented skin grafts. Bums., 1997, Nov – Dec; 23 (7-8): pp. 641-4.

8. Nelson – J.S.; Drenstein – A.; Liaw – L.H. & Berus, M.W.,(1996)., Mid-infrared erbium- YAG laser ablation of bone, the effect of laser osteotomy on bone healing . Laser-Surg-Med., 9(4): pp.362-374.
9. Snell – R., (2000). Clinical anatomy for medical students, 6th. Edition, Chapter (1): Introduction, pp. 35, Copy right 2000 Lippincott williams and wilkns.
10. Ihsan, M.F.R ; Effect of low level intravascular blood irradiation on the level of the genetamicin in the plasma. Photodiagnosis & Photomedicine Therapy, Volume 5, Issue; supplement 1, Pp; S36, 2008.
11. Aleksandrov M.T.,(199The working scheme of biological & theraputic effects of the laser irradiation. 1st – Scientific and practical conference of the Russian State Medical University Proceeding, Dec. (1992). pp. 1-6.
12. Golovin - S., (1992). Mechanism of laser action. 1st. - Scientific and practical conference of the Russian State Medical University Proceeding, Dec. (1992). pp. 7-9.
13. Ghamsari - SM.; Taguchi, K.; Abe, N; Acorda, JA. & Yamada, H., (1996). Histopathological effect of low-level laser therapy on sutured wounds of the teat in dairy cattle. et - Q, 1996, Mar; 18 (1): pp.17-21.