

Neocollagenesis in Deep and Superficial Dermis by Combining Fractionated Q-Switched ND:YAG 1,064-nm With Topical Plant Stem Cell Extract and N-Acetyl Glucosamine: Open Case Series

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ABSTRACT

Introduction: As we enter a new age of increasing demand in novel cosmetic therapies, we are challenged to provide excellent results with minimal downtime and safety in all skin types. In this open case series we are studying the improvement in rhytides by combining a novel, FDA-approved, non-ablative fractionated Q-switched ND: YAG 1,064-nm laser that acts in the deep dermis, with a topical containing plant stem cell extract and N-acetyl glucosamine (NAG) that acts in the superficial dermis.

Method: Six healthy females (Skin types III - V) were selected for the study with mean average age of 56 years +/- 11 years. The rhytides on the face and neck were assessed using a comprehensive grading scale. Patients were then divided into two groups, one received only laser treatment with the fractionated QSW 1,064 nm laser and the other group received combined treatment with the laser and topical. Patients were assessed again at 4 and 8 weeks.

Results: We observed an enhanced anti-aging effect of the laser in the patients with combined treatment.

Discussion: Understanding the effect of this novel laser therapy on human stem cells and investigating the basis of its synergistic effect with plant stem cell extract and NAG will lead us to better understand stem cell activity. Non-ablative tissue regeneration is the next step in providing optimal anti-aging treatments.

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INTRODUCTION

The demand for noninvasive skin rejuvenation is at the forefront of cosmetic medicine. As we strive to provide optimal Anti-Aging results with new medical procedure treatments, safety, pain, and downtime are important factors to consider. Photothermolysis with Fractionated laser devices has proven to provide neocollagenesis. These new age devices provide us with the benefits of no downtime and no pain.^{1, 2} The fractionated Q-switched ND: YAG 1,064-nm by Harmony XL, Alma Lasers Ltd. is the latest of these devices that provide us with the combination of safety, no pain, and no downtime. By keeping the skin intact, we avoid unnecessary delay in wound healing and risk of scarring but still have the dermal effects of collagen remodeling. Stem cell research is one of the most exciting topics in modern day biomedical investigation and stands vanguard of a new field called regenerative medicine. The enthusiasm about stem cell research derives from new insight into their behavior and location in the human body and from data that support the hope that stem cells may one day be used to repair injury in human tissues.³ The concept of scar less wound healing³ and the role of stem cells in collagen regeneration open the door to exciting possibilities in skin rejuvenation.

In this study we present an open series to compare the treatment of aging skin using fractionated Q-switched 1,064 with and without topical cream formulation containing plant stem extract having epigenetic properties to human stem cells and N-acetyl glucosamine (NAG) with a pro-collagen effect.

METHOD

Six women in the age group of 45-72 were selected in this study, with Fitzpatrick skin types III-V. They had visible signs of facial and neck skin aging. Verbal and written consents for treatment and photographs were obtained. The subjects were in good health with no preexisting skin conditions and were not on oral medications that could affect the laser treatment to the skin. Females that had prior use of fillers/botulinum injections or prior laser skin resurfacing procedures were excluded.

Scoring was based on rhytides, laxity, elastosis, dyschromia, erythema-telangiectasia, keratosis and texture. The Quantitative Comprehensive Grading Scale of Rhytides Laxity and Photoaging™ was used to grade each subject.⁴ Pretreatment photographs were taken at baseline.

TABLE 1.**Group A: Q Switched ND-YAG 1,064 Treatment No Topical**

Skin Type	Age	0 Weeks	4 Weeks	8 Weeks
V	72	3.5	3	2.5
IV	55	3	2.5	2.5
III	40	2.5	2.5	2
Mean	55.6	3	2.6	2.3
SD	16	0.5	0.28	0.28

TABLE 2.**Group B : Q Switched ND-YAG 1,064 With Topical**

Skin Type	Age	0 Weeks	4 Weeks	8 Weeks
V	65	3.5	3	2
IV	56	3	3	2.5
III	48	3	2.5	2
Mean	56.3	3.1	2.8	2.1
SD	8.5	0.28	0.28	0.28

The subjects were then divided into two groups, Group A that would receive only the laser treatment with fractionated Q-switched 1,064 nm and Group B that would receive combination of laser treatment and the topical cream.

Two treatments with the laser were done, one at baseline and then at 4 weeks. The group of subjects using the topical applied the cream daily to the face and neck throughout the course of the treatment for 8 total weeks. Patients were graded and photographs taken at 0-, 4-, and at 8-week intervals.

"It has been well-established that the tetracycline class of antibiotics may cause a variety of photo-induced drug eruptions."

The endpoint of treatment with the laser was mild erythema of the skin. Pain tolerance was assessed during and after the procedure. The energy settings for the fractionated Q-switched 1,064 nm ranged from 400-1200 mJ/pulse. The pulse repetition of the QSW is adjustable 1Hz, 2Hz, or 4Hz. Settings were adjusted based on patient's skin type. QSW laser suggested treatment parameters were used.²

RESULTS

We noted a slightly better response in overall texture, pigment, and tone improvement in Group B (laser and topical). We also note an enhanced response in the higher Fitzpatrick skin types

in both groups. The subjects that received topical also noticed an enhanced response to clearing of solar lentigines.

DISCUSSION

As technology and science advance we are constantly in pursuit of less invasive means to achieve optimal results. The desire for treatment options that minimize downtime and adjust to our patients' life style has top consideration. The ability to treat a growing mix of ethnicities safely in our patient population is also an important concern. New laser technology to manage aging skin warrants safety and effectiveness in all skin types.

The laser we have used in the study is the novel fractional, non-ablative Q-switched neodymium-doped YAG (ND: YAG) 1,064-nm laser (Harmony XL, Alma Lasers Ltd.).

Recently published studies on the Q-switched 1,064 nm ND:YAG laser device treating rhytides of face and neck reported improvement in photo aging skin, effectiveness in treating superficial rhytides, safety, and no downtime or pain.^{1,2} The mechanism of action of the fractional Q-switched 1,064 although not completely elucidated has to do with the fact that there are dermal heat zones created with the epidermis intact.^{1,5} This "photomechanical" effect leads to shrinkage of the Extracellular matrix (ECM)^{6,8} which in turn causes a stretching of the fibroblast. This change in the fibroblast shape then triggers an intracellular integrin mediated response that regulates increase in Tissue Growth Factor (TGF) and mRNA of procollagen enzymes, leading to collagen production. Dermal matrix in adult skin is composed of collagen type I (80-85%) and type III (10-15%), in addition to glycosami-

FIGURE 1.

FIGURE 2.

noglycan's and elastic fibers. Reduction of collagen type I and III are characteristics of chronologically aged skin and is also seen in photo damage. During the aging process collagen type III is gradually replaced by collagen type I. Therefore collagen type III is considered very important in skin collagen remodelling.⁸

Liu et al in 2008 reported that Q-switched ND:YAG laser has a photomechanical effect that promotes the synthesis of collagen type III, which is the predominant collagen in youth skin and therefore is more effective in treating elasticity of the skin.⁶ Dang et al 2010 studied in-vitro effects of the Q-switched 1,064 on human fibroblast and reported an increase in tissue growth factor beta and tissue inhibitors of matrix metalloproteinase (TIMP). The TIMP in turn reduce breakdown of collagen by inhibiting matrix metalloproteinase (MMP) in the extracellular matrix (ECM).^{5,6}

Histology done on patients post Q-switched 1,064 showed the most changes 2-3mm into the extracellular matrix of the upper reticular and papillary dermis. Also significant tissue response was noted in the para-adnexal areas.⁷

Stem cells are the most important cells in the skin. They are the source of continuous regeneration of the epidermis and the formation of new cells. In the human skin the stem cells are located in "niches" that are found in the dermo-epidermal junction bordering the papillary and reticular dermis and stratum basalis around the bulb of the hair follicle.⁹ Reduced viability and senescence of stem cell is a principal cause of tissue aging. Senescence is a natural process that after about 40 divisions stops the capacity of the cell to undergo further divisions. But senescence can also happen earlier in a cell's life, for example as a response to damage of cellular DNA. Premature damage is

especially detrimental when it hits stem cells because they are indispensable for tissue regeneration.¹⁰

This is an open case series to report the combined effect of the non-ablative fractionated Q-switched ND: YAG 1,064 with a topical. The topical cream contains apple stem cell extract and N' Acetyl glucosamine, that have pro-collagen and stem cell regenerative effects (Neostrata Inc.).

We noticed that the group of patient that had a combined treatment showed a slightly better improvement in overall rhytides pigment change as compared to the group that just had the laser treatment.

The apple stem cell extract is derived from the Uttwiler Spätauber apples. These apples are known to stay fresh for an extraordinary long time. The extract obtained was used in vitro studies and showed reduction of fibroblast senescence. In vivo topical application in the form of PhytoCellTec Malus Domestica cream has shown to cause reduction in rhytides. The epigenetic properties of the apple stem cell extracts are shown to have antioxidant effects on human fibroblast and a possible nutritive effect on the human stem cell.¹⁰⁻¹²

N-acetyl glucosamine (NAG) has recently gained a lot of excitement in the cosmetic science community, showing its effects on increasing fibroblastic activity in in-vitro studies and causing increases in gene regulation of sugar metabolism in fibroblast. It also is one of the important building blocks of hyaluronic acid,

FIGURE 3.

FIGURE 4.

which is a part of the extracellular matrix. It has shown to reduce inflammation by upregulating antioxidant enzymes and enzymes that regulate sugar metabolism (DL Bisset et al). NAG also has a tyrosinase inhibitor effect helping with pigment reduction. The effects of topical NAG are discussed by Schlesinger et al, 2015, particularly in improvement of neck rhytides.¹³⁻¹⁵

We report the first case series done combining fractionated Q-switched 1,064 nm with a topical plant stem cell extract and NAG. The improvement in patients that received a combined treatment can be explained by the different depths of action of the respective treatments.

The area of action of the fractionated Q-switched 1,064 in the upper reticular dermis, about 2-3 mm depth, with adnexal changes (Paasch et al) around the site of the epidermal stem cell 'niches'. The topical is acting at the epidermal and papillary dermal layers 1-2 mm deep. The area of synergy is at the dermo-epidermal junction and papillary dermis.

We also propose that the Q-switched 1,064 nm wavelength action on the ECM and fibroblast activity causes increased activity of procollagen enzymes and thus increases demand of building blocks of collagen, which the topical NAG provides. Apple stem extract also provides anti-oxidant properties that help with the process of sugar metabolism.

We still have to explore the exact effects of the laser on the human stem cell "niche" and further histological studies with collagen mapping will help us understand the exact effect of the laser on fibroblast, human stem cells, and ECM. Further

studies combining treatment with topical plant stem cell extract and NAG with histological and biochemical data will help us understand the exact mechanism of the synergistic effect.

Our study also notes a more significant response to Q-switched 1,064 nm in darker skin types in both groups. This also opens the idea to explore histological changes in darker skin with Q-switched 1,064 laser and if there is a predominance of certain collagen type in these groups that leads us to have different aging patterns.

We are at the forefront of a new age that needs to explore safe, no downtime laser treatments in combination with effective topicals. Stem cells are the most important cells in the skin. Their unique and special properties help in regeneration of the epidermal and dermal layers of the skin. Stem cell research holds the key to understanding aging patterns and may give us the tools we need to modify aging. Identifying the effects of topicals and laser procedures on the stem cell "niches" that are found in the epidermal layer should be our next step in understanding their unique physiology.

CONCLUSION

The excellent *in vivo* results of this open series challenge us to further explore the synergistic effects of these novel modalities *in vitro*. Tissue regeneration is the new frontier of cosmetic medicine opening new avenues for scar healing, wound repair, and anti-aging.

DISCLOSURES

The authors have no conflicts of interest to declare.

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