

USING LED PHOTOBIMODULATION TO TREAT PREMATURE AGEING

Leonardo Marini, Giuliana Crisman, Vera Trashlieva, Aleksandar Krunic, Panagis Polizois and Andrea De Faveri present the findings from a clinical study on the sequential use of continuous emission and pulsed emission light therapy

ABSTRACT

Background and objective

Facial skin shows signs of ageing earlier than other anatomical areas. Predominantly non-thermal infrared A (IR-A) light-emitting diode (LED) photobiomodulation has proven effective in triggering intracellular photobiochemical reactions leading to new collagen synthesis and reduction of matrix metalloproteinase-1 (MMP-1). The objective of this study was to assess the effectiveness, safety, and tolerability of a sequentially combined, continuous (CW) 835±5 nm and pulsed emission (PW) 875±5 nm LED facial mask array in the treatment of facial premature ageing.

Materials and methods

Ten female subjects (47-77 years; mean 55 years) affected by photoageing were

enrolled in this pilot prospective single cohort study. Facial skin was exposed to a sequentially combined 840 nm and 880 nm LED for 15 minutes twice per week for 4 weeks. Elastometry, skin colourimetry, and standardised computerised facial complexion analysis were performed to assess clinical improvements. Standardised digital photographs were taken immediately pre- and 45 days post-treatment to evaluate the internal consistency between two independent blinded observers who were asked to sort out and identify pre- and post-treatment images.

Results

Skin texture, wrinkle count, and elasticity were remarkably improved 15 days and 45 days after eight LED facial treatments. Surface erythema showed a moderate increase after the same intervals of time.

Minimal surface micro-dyspigmentation was detected by computerised digital skin complexion analysis 15 days and 45 days after eight LED treatments. Skin texture, tone, and wrinkles were considered improved by the majority of subjects. Moderate burning and warmth sensation were reported by 70% of subjects during LED exposures. Identification of pre-treatment digital images of treated subjects by two independent, blind dermatologists showed a 90% concordance.

Conclusions

A series of combined, sequential CW 840 nm and PW 880 nm narrowband LED irradiations, delivered through an innovative anatomically designed facial mask, has proven effective in temporarily improving premature ageing-related structural and functional skin alterations.



LEONARDO MARINI, MD, is Medical Director at The Skin Doctors' Center, Italy; **GIULIANA CRISMAN, MD**, is a Consultant Dermatopathologist at The Skin Doctors' Center; **VERA TRASHLIEVA, MD**, is a Senior Consultant at The Skin Doctors' Center; **ALEKSANDAR KRUNIC MD**, is Associate Professor of Dermatology at University of Illinois, Chicago, and a Consultant at The Skin Doctors' Center; **PANAGIS POLIZOIS MD**; Consultant Dermatologist at The Skin Doctors' Center; **ANDREA DE FAVERI** Statistician at The Skin Doctors' Center
email: leonardo.marini@skindoctors.it

KEYWORDS

collagen remodelling, LED photobiomodulation, photorejuvenation

PREMATURE SKIN AGEING IS more pronounced on sun-exposed body areas, particularly on facial regions, where intrinsic, environmental, and lifestyle factors are intimately associated in determining typical morphological changes, including wrinkles, telangiectasia, dryness, rough texture, yellowish colour plaque-like thickening, and pigment irregularities.

Collagen synthesis is progressively reduced, while interstitial matrix metalloproteinases (MMP; MMP-1 in particular) are up-regulated in aged skin^{1,2,3}. It has been demonstrated that a variety of narrow band, incoherent light-emitting diodes (LEDs) can stimulate natural intracellular photobiochemical reactions, producing clinical improvements of aged skin as well as accelerating wound repair through predominantly non-thermal photobiological processes^{4,9}. Karu and Kolyakov identified four 'active' regions in the wavelength range considered important for phototherapy (600-860 nm), after analysing action spectra of red infrared (IR) (580-860 nm) monochromatic light; two were located in the red range (613-623 nm and 667-683 nm) and two in the IR range (750-772 nm and 812-846 nm)¹⁰. Claimed

photobiological effects have included fibroblasts activation, improved neovascularisation, enhanced neocollagenesis, and macrophage biostimulation, mostly induced after light absorption by cellular photoacceptors^{11,2}.

Continuous emission (CW) has been considered as the gold standard for the majority of low level light therapy (LLLT); however, some studies indicate that pulsed light emission (PW) may be superior to CW keeping constant all other parameters¹³. Different pulse frequencies have been used, but there is no universal consensus on the identification of the most effective¹⁴⁻²¹. All previously listed biological effects are quite different from those produced by molecular 'broadband IR response' seen after prolonged exposure to IR radiation (760 nm-1 cm). The IR spectral region is arbitrarily divided according to wavelength into three sub-regions: IR-A (760-1400 nm); IR-B (1400-3000 nm); IR-C (3000 nm-1 mm). IR constitutes 54.3% of total solar irradiance, while IR-A accounts for approximately one third of solar energy load of human skin. The depth of penetration into skin and subcutaneous tissue decreases with increasing wavelength in the IR spectrum. Broadband IR radiation typically induces molecular vibrations and rotations, >

A woman's profile is shown in a serene, meditative pose with her eyes closed. She is surrounded by a soft, ethereal glow of blue light particles and bokeh effects, suggesting a therapeutic or healing environment. The background is a deep, dark blue, making the lighter blue particles stand out prominently.

“ Continuous emission has been considered as the gold standard for the majority of low level light therapy; however, some studies indicate that pulsed light emission may be superior. ”



Figure 1 LED facial mask and EPI-C Plus main unit used in this study

▷ causing increased tissue temperature as well as generating complex biochemical cellular effects.

Considering the different photobiological tissue response observed after narrow-band IR-A incoherent LED and broadband IR-A incoherent light, the importance of producing a selective fragmentation of IR-A to separate appropriate IR-A wavelengths able to induce positive, predominantly non-thermal, photobiochemical reactions eventually leading to clinical improvement of skin function and appearance is quite evident. This article presents the results of a pilot clinical study involving a group of 10 individuals affected by premature ageing (chrono- and photoageing) treated by a novel narrow band, sequential CW and PW light delivery system produced by a sequentially combined 840nm CW followed by 880nm PW LED array, innovatively distributed along the inner surface of an anatomically designed polystyrene facial mask.

Materials and methods

This study investigated the effectiveness of the EPI-C Plus main unit (Espansione Marketing Spa, Italy), an innovative 840-880nm LED facial mask developed to induce skin photobiomodulation (Figure 1) and cleared by the Food and Drug Administration (FDA). Treatment protocol was also approved by the Continuous Dermatologic

Figure 3 Clinical pictures taken at T0 (immediately pre-treatment), T45 (15 days after eight LED sessions), and T75 (45 days after eight LED sessions)

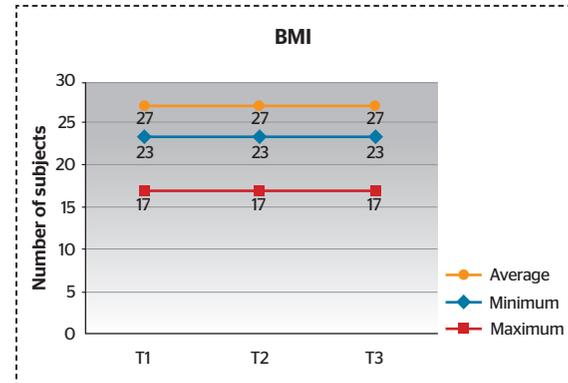


Figure 2 Body mass index of treated subjects, showing a constant stability of values for the duration of the study

“ Weight gain could sensitively decrease lines and wrinkles on the face, thus leading to an important bias in this kind of pilot study. ”

Education Committee (CDEC) Review Board (Trieste, Italy), and comprises a sequential combination of CW 835±5nm followed by PW narrow-band 875±5nm LED light facial mask for the treatment of premature ageing.

Subjects

Ten healthy female subjects with premature ageing with no previously performed invasive and/or non-invasive anti-ageing procedures, were recruited between March and April 2011 according to a protocol approved by the CACD Review Board. Ages ranged from 47 to 77 years with a median age of 55 years. Inclusion criteria were:

- Fitzpatrick skin types I-III presenting with mild-to-moderate facial premature ageing (score 1.5-2.5 on modified Fitzpatrick wrinkle scale)²²
- Positively oriented psychological attitude to improve skin appearance and function
- Realistic expectations concerning clinical results after mild, non-invasive procedures.

Exclusion criteria included:

- Pregnancy
- Important facial scarring from previous inflammatory and/or traumatic diseases
- Recent sunburns
- Topical anti-ageing treatments within 6 months prior to treatment
- Laser/IPL
- Chemical peel
- Cosmetic facial surgery within 12 months prior to treatment.

Full patient consent to participate in the study was obtained using an IR-B approved protocol and consent form. Body mass index (BMI) was calculated during all scheduled clinical appointments for each subject in order to monitor the stability of their general metabolism for the duration of the study. In fact, weight gain could sensitively decrease lines and wrinkles on ▷

Figure 4 Computerised facial complexion analysis (wrinkles) at T0 (immediately pre-treatment (A)) and T45 (15 days after eight LED treatments (B))



“All ten subjects completed the full series of treatment sessions and attended all scheduled follow-up visits.”

▷ the face, thus leading to an important bias in this kind of pilot study.

Treatment

All subjects were asked to temporarily suspend routinely applied topical facial products 15 days prior to initiation of treatment. A standardised skincare regimen was prescribed during the 15-day pre-treatment ‘wash-out period’ according to the following:

- Ictyane cleansing cream (Ducray Laboratoires Dermatologique, Pierre Fabre, France) – one application twice daily
- Thermal spring water mist (Avène, Pierre Fabre, France) – one mist twice daily immediately before moisturising cream
- Ictyane moisturising cream (Ducray Laboratoires Dermatologique, Pierre Fabre, France) – one application twice daily immediately after Thermal spring water
- Anthelios SPF 50+ cream (La Roche Posay Laboratoires Dermatologique, France) – one application every morning on top of moisturising cream, to be repeated every 2 hours in cases of prolonged UV exposure.

The same skincare regimen was maintained for the duration of LED treatment, and represents a standard hydrating protocol with creams with hydrating properties and none with anti-ageing properties. This standardisation was introduced to avoid any bias from other anti-ageing creams eventually applied subjectively at home.

All patients received eight full-face LED treatments over a 1-month period. Two treatments were scheduled per week, with a minimal interval of 3 days between treatments. All patients were instructed not to wear any facial make-up from the night prior to treatment. Gentle facial cleansing with Ictyane cleansing cream was performed immediately pre-LED light exposure. Cleanser was rinsed off with lukewarm water. The LED facial mask was carefully positioned on facial skin and retaining straps were adjusted to obtain a perfect anatomical match between LED array and facial skin, keeping light source distance to an average of 5mm (8±4mm). Each treatment consisted of standardised 15minute pre-set 835±5nm CW LED (50%) plus 15minute pre-set 875±5nm PW LED (50%) exposures for a total irradiation dose of 66J/cm² per session (33J/cm² + 33J/cm²).

Immediately post-treatment, Thermal spring water mist followed by Ictyane moisturising cream were applied to treated skin. All subjects were instructed to wear daily high SPF sunscreen, and minimise sun exposure. Make-up was allowed 1 hour after treatment.

Clinical photographs

Standardised clinical photographs were taken at baseline, immediately before the first LED exposure (T1), at 15 days (T2-T1+45days), and 45 days (T3-T1+75days) after completion of eight treatment sessions using a 10.5 megapixel EOS 1000D digital camera (Canon Inc, Tokyo, Japan), a EF-50mm f/2.5 Compact-Macro lens (Canon Inc.), and a 220EX Speedlite dedicated TTL flash ▷

Figure 5 Skin texture digital features count variations (standardised L + R cheek Visia complexion analysis measurements – median values) and corresponding percentages of global texture improvement (L + R cheek – median values) at T1=baseline, T2=15 days, and T3=45 days post-treatment

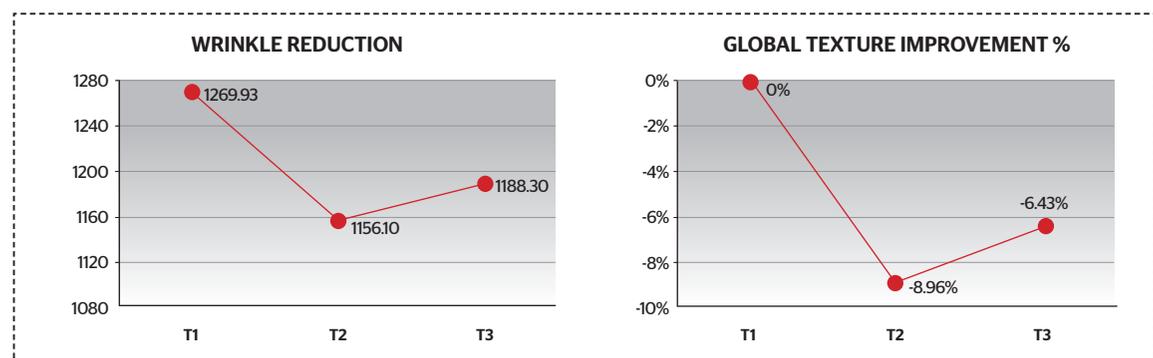
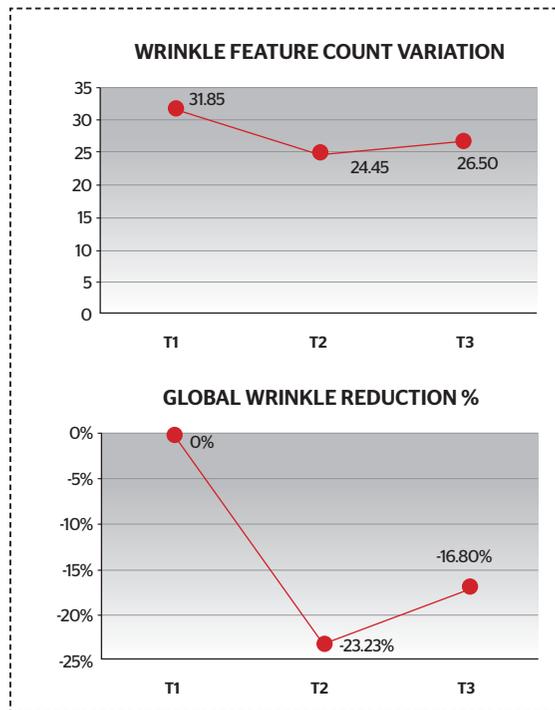


Figure 6 Skin wrinkles digital features count variations (standardised L+R cheek Visia complexion analysis measurements—median values) and corresponding percentages of global wrinkles reduction (L+R cheek—median values) at T1=baseline, T2=15 days, and T3=45 days post-treatment



“ Subjective assessment of clinical results were obtained 15 days and 45 days after completion of full treatment sessions. ”

▷ (Canon Inc.). In the aim to standardise templates, all subjects were asked to wash off any make-up before photographic sessions and rest sitting for 15 minutes at constant thermal comfort room parameters: temperature (23.5-25.5°C) and relative humidity (50±5%). At the end of each photographic session an *in vivo* 3D photographic quantitative measurement of skin texture, wrinkles and superficial pigmented alterations was obtained using a Visia Complexion Analysis standardised photographic system with Visia 5 software (Canfield Imaging System, Canfield Scientific, NJ, USA).

Erythema and elastometry

Skin erythema was measured using a II ColorMeter (Cortex Technology, Denmark). Elastometry values were measured using a Dermacheck Multi Skin Center MC750 (Courage-Khazaka Electronic GmbH Kolin, Germany). All measurements were taken on standardised facial anatomical landmarks identified by the intersection between a vertical line passing through the centre of the pupil and a horizontal line touching the base of the columella on both cheeks. Erythema was

measured first in order to avoid possible secondary vaso-reactivity induced by the suction probe used to measure elastometry. All measurements were taken at constant thermal comfort room parameters, immediately before the first treatment session (T1), and subsequently at 45 days (T2-T1+45 days) and 75 days (T3-T1+75 days).

Subject's self-assessment

Simple questionnaires to evaluate subjective intra-operative perceived sensations were given to each subject at the end of the first treatment (T1-IOP) and at the end of the full eight treatment sessions (T1+30 days-IOP). Seven parameters were evaluated during LED exposure:

- Pleasant feeling (PF)
- Absence of pain (AOP)
- Safety perception (SP)
- Absence of fear (AOF)
- Burning perception (BP)
- Warmth perception (WP)
- Claustrophobia perception (CP).

Four different sensations were evaluated immediately after the end of the first treatment (T1-END) and at the end of the complete treatment sessions (T1+30 days-END): pleasant feeling (PF); absence of pain (AOP); burning perception (BP); and warmth perception (WP). Five more subjective perceptions were evaluated 24 hours after the first treatment (T1-END+24 hours) and after the complete treatment sessions (T1+30 days+24 hours):

- Not interfering with social activities (NISA)
- Not interfering with make-up use (NIMU)
- Absence of pain (AOP)
- Absence of desquamation (AOD)
- Absence of redness (AOR).

Each parameter was evaluated according to a five-point scale as follows: 1=totally disagree, 2=disagree, 3=indifferent, 4=partially agree, 5=completely agree.

Subjective assessment of clinical results were obtained 15 days (T2-T1+45 days) and 45 days after completion of full treatment sessions (T3-T1+75 days). Study subjects were previously instructed on how to evaluate their skin tone improvement, wrinkle reduction, colour uniformity improvement, and skin texture improvement; thus, all parameters were subjectively evaluated on a five-point assessment scale as follows: 1=totally ineffective, 2=moderately effective, 3=effective, 4=very effective, and 5=extremely effective. ▷

Figure 7 Skin dyspigmentation digital features count (standardised L+R cheek Visia complexion analysis brown spots measurements—median values) and corresponding global skin dyspigmentation percentage variations (L+R cheek—median values) at T1=baseline, T2=15 days, and T3=45 days post-treatment

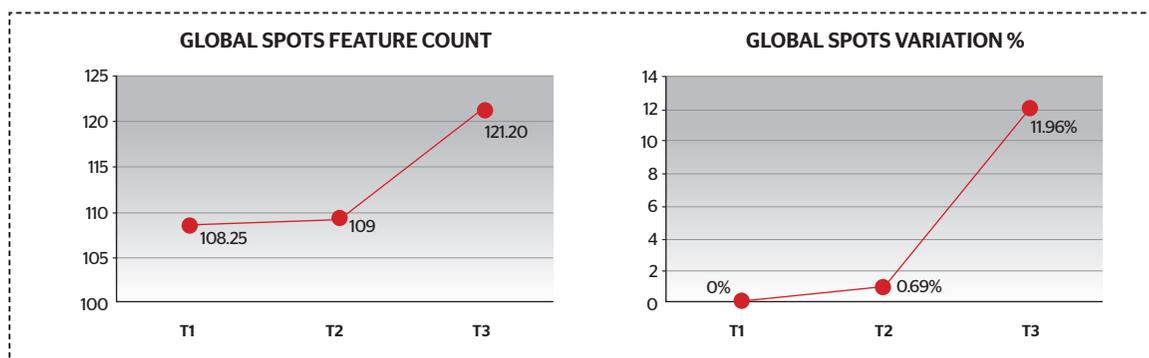
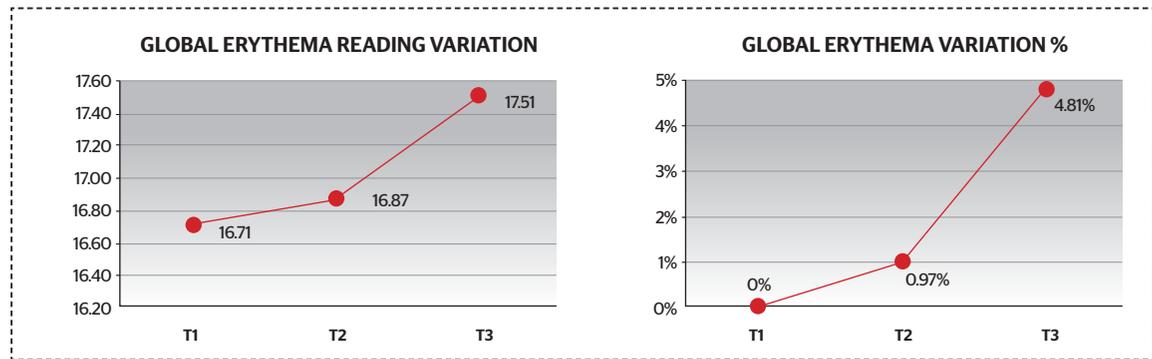


Figure 8 Skin erythema global value variations (standardised L + R cheek spot measurements – median values) and corresponding percentages of global elastometry improvement (L + R cheek – median values) at T1=baseline, T2=15 days, and T3=45 days post-treatment



▷ **Overall clinical outcome assessment**

Clinical photography assessment was performed blindly by two independent dermatologists who were asked to identify the temporal order of digital pictures taken at T1: immediately pre-treatment; T2: T1+45 days (15 days after the completion of eight LED treatments); and T3: T1+75 days (45 days after the completion of eight LED treatments), after being blindly reshuffled by a nurse. A simple percentage match was used to evaluate the magnitude of agreement between the two reviewers.

Results

All ten subjects completed the full series of treatment sessions and attended all scheduled follow-up visits. No variations were detected regarding subjects' BMI during the duration of the study (Figure 2).

Overall clinical outcome photographic assessment by independent reviewers

Both reviewers correctly identified the pre-treatment photographs (100% concordance). Moderate disagreement was registered with regard to the correct assignment of T2 (T1+45 days) and T3 (T1+75 days) images (72% concordance) (Figure 3).

Objective assessment of skin texture, wrinkles, and superficial pigment alterations

Skin texture irregularities, wrinkle count, and superficial pigment irregularities were automatically evaluated using the computerised feature count system installed on a Visia Complexion Analysis unit. Computerised assessments were made immediately before the first LED treatment (T1), at T2 (T1+45 days), and at T3 (T1+75 days).

Global skin texture feature count was found significantly decreased from baseline values at T2 (-8.96%) (Figure 4) followed by a slight upward variation, keeping values still below baseline at T3 (-6.93) (Figure 5). Global wrinkle feature count showed an even more marked decrease from baseline values at T2 (-23.23%), followed by a slight upward variation at T3 (-16.8) (Figure 6). Superficial spot feature count showed an opposite global trend scoring a +0.69% at T2 and a +11.96% at T3 (Figure 7).

Erythema and elastometry

Surface skin erythema showed a slight increment at T2 (+0.97%) followed by a more pronounced increase at T3

(+4.81%), confirming the positive metabolic stimulation of LED treated regions (Figure 8). Global elastometry showed a marked improvement at T2 (+20.59%) followed by a moderate decrease (+10.99%), keeping still above baseline values, at T3 (Figure 9).

Subject's self-assessment

Study subjects were asked to evaluate their intra-operative perceptions (IOP) immediately after their first LED treatment (T1-IOP) and immediately after their last LED session (T1+30 days IOP). Intra-operative treatment acceptability was considered globally good regarding all assessed subjective sensations except moderate claustrophobia (CP), slight superficial BP, and moderate skin WP, both reported immediately after the first treatment session (T1-IOP) and immediately after the last treatment session (T1+30 days-IOP). Moderate claustrophobic sensation was reported in 20% of subjects at (T1-IOP), and 30% at (T1+30 days IOP). Moderate burning sensation was reported by 40% of subjects both at T1-IOP and T1+30 days IOP. Similarly, 70% of treated patients experienced a warmth sensation both at T1-IOP and T1+30 days-IOP. Curiously, a slightly better intra-operative acceptability score regarding 'pleasant feeling' and 'absence of fear' was reported at T1+30 days, probably because study subjects experienced a progressive adaptation to intra-operative associated symptoms during the course of LED treatments. Of note, AOP was perceived by 100% of subjects (90% totally agree, 10% partially agree) both after the first treatment (T1-IOP) and at the end of treatment sessions (T1+30 days-IOP). Ninety percent of study subjects considered these treatments as high safety profile procedures (70% totally agree, 20% partially agree at T1-IOP; 60% totally agree, 30% partially agree at T1+30 days-IOP). Absence of fear was reported by 90% of subjects (80% totally agree, 10% partially agree) at T1-IOP, while 100% totally agree at T1+30 days-IOP. When asked to regard their sensations immediately after LED exposure study subjects reported a pleasant feeling in 40% of cases at T1-IOP and in 50% of cases at T1+30 days IOP (Figure 10).

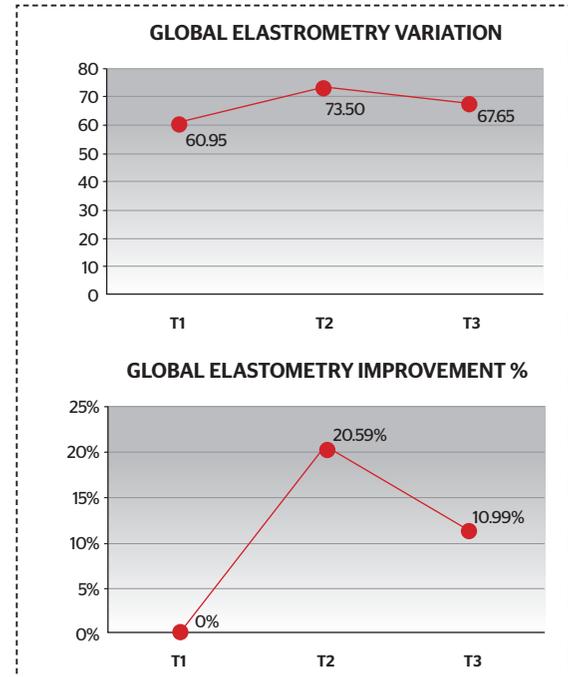
Study subjects were asked to evaluate the sensations perceived on their skin immediately after the first treatment session (T1-END) and immediately after the last treatment session (T1+30 days-END) regarding PF, AOP, BP, and WP. Forty percent of subjects regarded their

“Absence of pain was confirmed by 90% of subjects, while 10% were indifferent at the end of the first LED treatment.”

LED treatment as pleasant both at the end of the first session and at the end of the last session. Twenty percent of subjects considered the treatment unpleasant after the first session, while no subjects were of the same opinion after the last treatment session. Eighty percent of subjects reported a complete absence of pain, while 20% were totally indifferent both at T1-END and at T1+30 days-END. With regard to BP, 20% of subjects reported a mild burning sensation immediately post-treatment at T1-END, while 40% confirmed this sensation at T+30 days-END. IR photo-thermal burning is usually associated with a perception of deep tissue warmth and this was confirmed by 50% of subjects at T1-END and 70% at T1+30 days-END (Figure 11).

Study subjects were further asked to evaluate the superficial perception of their skin 24 hours after the first treatment (T1-END+24 hours) and 24 hours after the last LED session (T1+30 days+24 hours). One hundred percent of subjects totally agreed with regard to the absence of procedure-related interferences with their usual social activities (NISA=not interfering with social activities) at T1-END+24 hours, while 90% of subjects confirmed their initial evaluation, and 10% partially agreed to it at the end of the last LED treatment (T1+30 days+24 hours). Regarding the possibility of applying their usual make-up after the procedure (NIMU = not interfering with make-up use), 90% of subjects totally agreed about the absence of any interferences

Figure 9 Skin elastometry global value variations (standardised L+R cheek spot measurements—median values) and corresponding percentages of global elastometry improvement (L+R cheek—median values) at T1=baseline, T2=15 days, and T3=45 days post-treatment



induced by LED irradiation, while only 10% partially agreed with this point at T1-END+24 hours. Ten percent of subjects disagreed with this, while 80% totally agreed and 10% partially agreed at the end of the last LED

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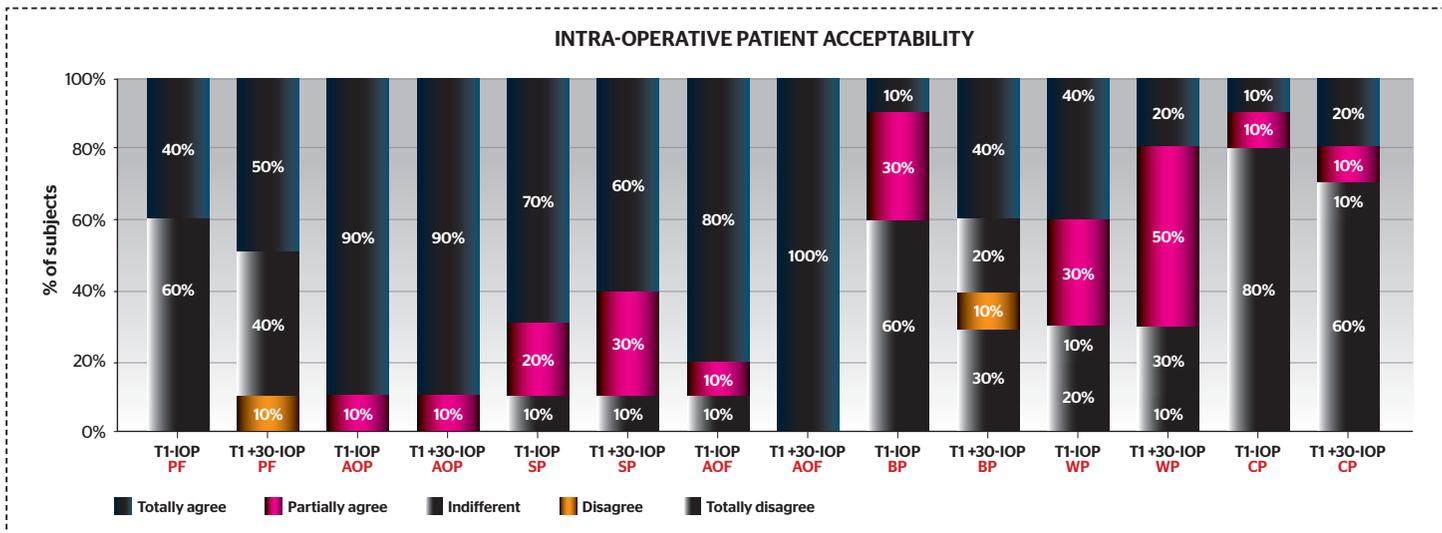


Figure 10 Subjective perception of intra-operative LED treatment acceptability as reported by study subjects immediately after the first session (T1-IOP) and immediately after the last treatment (T1+30-IOP): PF = Pleasant Feeling; AOP = Absence of Pain; SP = Safety Perception; AOF = Absence of Fear; BP = Burning Perception; WP = Warmth Perception; CP = Claustrophobia Perception

▷ session (T1+30 days+24 hours). Absence of pain was confirmed by 90% of subjects, while 10% were indifferent at the end of the first LED treatment (T1-END+24 hours). One hundred percent of patients agreed with this perception (90% totally, 10% partially by referring a mild BP) at the end of the last treatment (T1+30 days+24 hours). Absence of superficial desquamation (AOD) was perceived by 80% (totally agree) of subjects, while 10% disagreed, and only 10% totally disagreed at the end of the first LED treatment (T1-END+24 hours). With regard to the same observation, 90% of subjects totally agreed and 10% confirmed their indifference at the end of the last LED session (T1+30 days+24 hours). Absence of post-treatment redness (AOR) was confirmed by 70% (totally agree) and 20% (partially agree), while only 10% totally disagreed with this observation at the end of the first LED treatment (T1-END+24 hours). Regarding the same point, 60% of subjects totally agreed and 30% partially agreed at the end of the last LED session

“Absence of pain was confirmed by 90% of subjects, while 10% were indifferent at the end of the first LED treatment.”

(T1-END+24 hours). The percentage of subjects who totally disagreed with this point remained the same (10%) (Figure 12).

Subjective assessment of clinical results confirmed what was already evident after the evaluation of objective parameters considered for the study. Skin texture improvement was considered quite significant by 100% of study subjects. Overall effectiveness reported at T2 was regarded as extremely effective (40%), very effective (30%), effective (20%), and moderately effective (10%). The same 100% effectiveness was also reported at T3 (30% extremely effective, 20% very effective, 20% effective, and 30% moderately effective). Ninety percent of subjects also considered the treatment effective in improving their wrinkles at T2. Of these, 20% considered the treatment extremely effective, 40% very effective, 20% effective, and 10% moderately effective. The same overall effectiveness percentage was registered at T3, with moderate inter-subject variations: 20% of subjects ▷

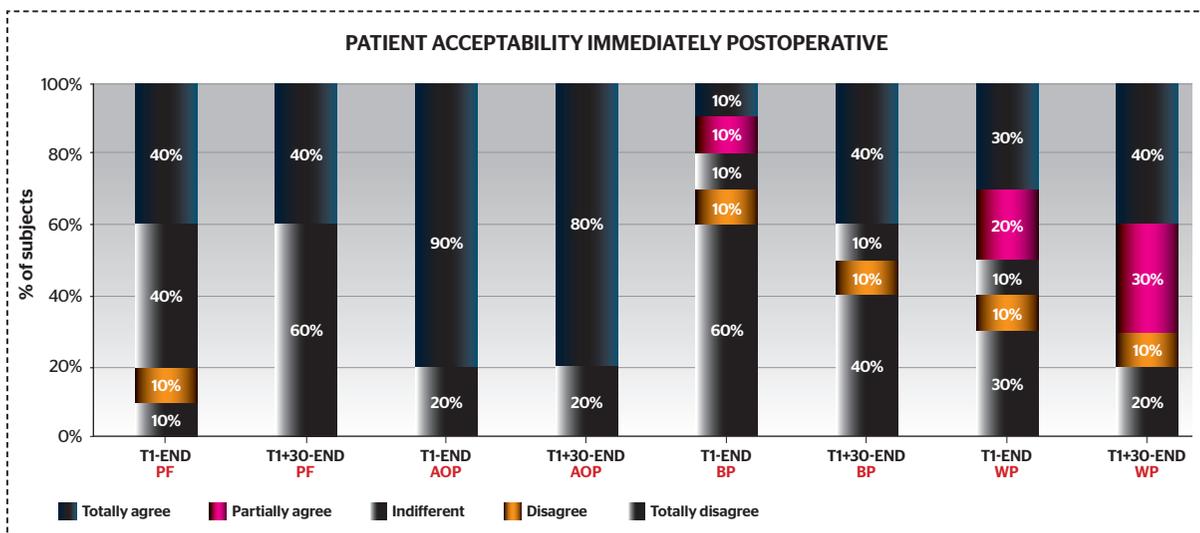
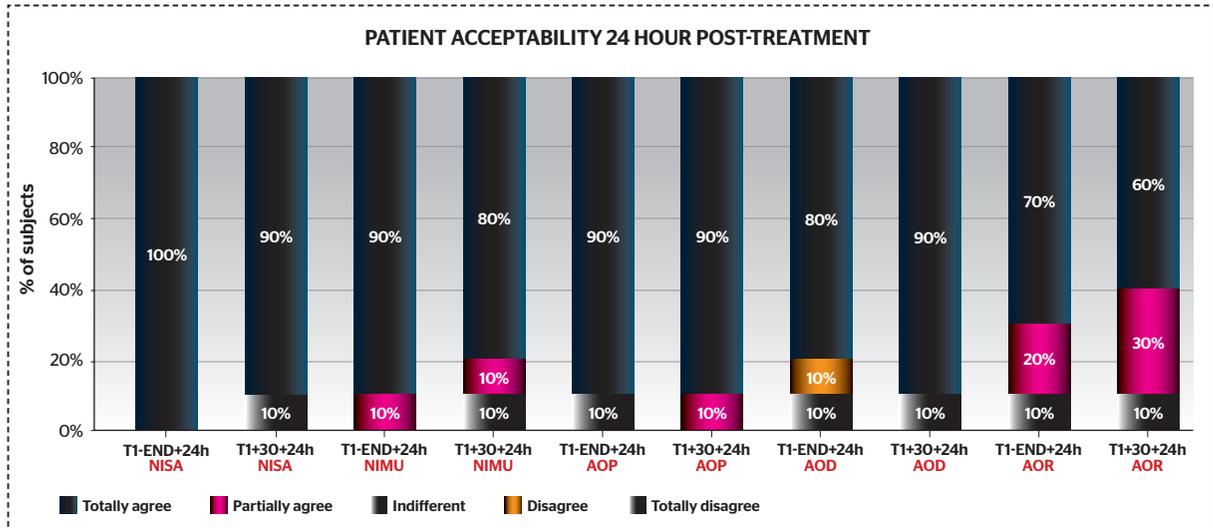


Figure 11 Subjective perception of immediate post-operative LED treatment acceptability as reported by study subjects after the first session (T1-IOP) and after the last treatment (T1+30-IOP): PF = Pleasant Feeling; AOP = Absence of Pain; BP = Burning Perception; WP = Warmth Perception

Figure 12 Subjective perception of late postoperative LED treatment acceptability (24 hours after treatment) as reported by study subjects after the first session (T1+OP) and after the last treatment (T1+30+OP):
 NISA = Treatment Not Interfering with Social Activities;
 NIMU = Treatment Not Interfering with use of Make-Up; AOP = Absence of Pain; AOD = Absence of Desquamation; AOR = Absence of Redness



▷ considered the treatment extremely effective, 20% very effective, 20% effective, and 30% moderately effective (Figure 13).

The most important improvement reported was on overall skin tone perceived by study subjects. All treated subjects considered LED photobiomodulation effective at T2 (T1+45 days). Fifty percent considered the treatment extremely effective, 40% very effective, and 10% moderately effective. Slight variations in the clinical evaluation score were observed at T3 (T1+75 days), always keeping a 100% satisfaction rate. Of these, 30% of subjects considered the treatment extremely effective, 20% very effective, 20% effective and 30% moderately effective. In spite of the opposite trend observed for superficial skin spots as measured by Visia computerised skin complexion analysis, most study subjects considered this specific parameter improved. Eighty percent of subjects confirmed an improvement at T2 (30% extremely effective, 20% very effective, 10% effective, and 20% moderately effective). Ninety percent of subjects confirmed an improvement also at T3 (30% extremely effective, 10% very effective, 30% effective, 20% moderately effective) (Figure 14).

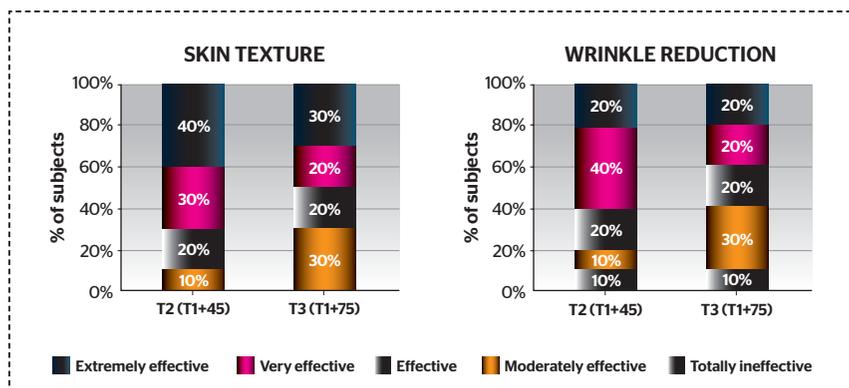
“The most important improvement reported was on overall skin tone perceived by study subjects.”

rejuvenate exposed skin by positively modulating overall cellular levels, increasing dermal collagen synthesis, and down-regulating matrix metalloproteinases. The final clinical result of LED photobiomodulation should therefore correlate with progressive dermal structural changes triggered by sequential exposures to specific wavelengths. Two major potential drawbacks of this interesting approach to skin rejuvenation are identified by the short temporal coherence (coherence length) and the low spatial or lateral coherence characterising narrowband LED, making treatment distance between light source and irradiated surface crucial for the effectiveness of photobiological events. The more monochromatic the light, the longer the length where the light field is coherent in volume, and the smaller the divergence of the light beam at the point of irradiation²³. These particular physical aspects of LED radiation allow for uniform irradiance on highly irregular skin surfaces, like the human face. Most LED delivery systems consist of linear stripes of variable length, regularly distributed along flat or curved panels. This feature showed to be only partially effective in ‘recontouring’ the complex anatomy of the human face.

Discussion

Narrowband incoherent IRA LED photobiomodulation within the action spectra of 580nm to 860nm should

Figure 13 Subjective perception of LED treatment efficacy on overall skin texture improvement and wrinkle reduction at T2 = T1+45 days, and T3 = T1+75 days as reported by study subjects



In the present study a novel narrowband sequentially combined 840nm CW+880nm PW LED facial mask light delivery system has proven to be safe and effective in reducing facial wrinkle count, improving overall skin texture, tone, and elasticity. A minor increase in surface spotty dyspigmentation count was detected by computerised facial feature analysis by Visia Complexion Analysis, 15 and 45 days after the treatment. This particular finding could be explained as a sub-clinical post-inflammatory hyperpigmentation (PIH) due to a micro-thermal paracrine-cytokine signaling up-regulation of keratinocyte and fibroblast alpha-Melanocyte Stimulating Hormone (MSH) release P53-dependent²⁴⁻²⁵. The majority of study subjects considered the treatment quite effective (90-100%) and acceptable, both intra-operatively, and post-operatively. The combination of CW emission immediately followed by PW emission may possibly explain the high degree of clinical success in improving premature facial skin ageing. ▷

▷ The initial part of LED irradiation, consisting of CW light emission, could have been associated with a photobiochemical and moderately photothermal 'priming' of exposed cells, preparing the ground for a better response to the on-off biostimulatory effect induced by PW irradiation during the final part of the treatment. The idea of evenly distributing LED arrays along the inner surface of an anatomically designed polystyrene facial mask has shown to be quite effective in keeping light delivery distance as short and constant as possible ($8\text{mm} \pm 4\text{mm}$), all along the complex surface anatomy of facial skin. This particular innovative feature could have been responsible for a consistent optimisation of facial irradiance and consequently, for the overall photobiological effectiveness of this treatment.

Small sample size and full-face treatment represent two study limitations, but split-face studies are already planned to further prove the efficacy of this interesting non-invasive skin rejuvenation protocol.

Conclusion

Predominantly non-thermal high-energy LED photobiomodulation performed with an innovative facial mask light delivery system using a sequential combination of 840nm CW and 880nm PW irradiation has proven effective in positively modifying skin tone and texture of prematurely aged facial skin. Clinical results were confirmed by objective and subjective assessment 15 and 45 days after completion of full treatment sessions. Textural, wrinkle, and skin elasticity modifications were found more pronounced 15 days after the end of the eight LED treatments and showed a slight decrease after 45 days, thus indicating the need for a periodical repetition to keep tissue functional improvements. Sequential combined narrow-band CW+PW LED treatment was reported to be painless, safe, and easy to perform.

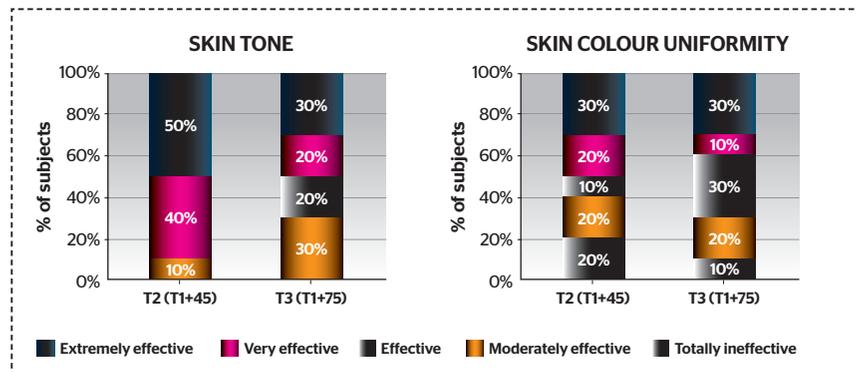


Figure 14 Subjective perception of LED treatment efficacy on overall skin tone improvement and skin colour uniformity at T2 = T1 + 45 days, and T3 = T1 + 75 days as reported by study subjects

The combination of such visible and consistent aesthetic improvement and the feasibility of the treatment sessions led all study subjects, in the end, to agree in recommending an anti-ageing treatment with EPI-C Plus 840–880nm LED facial mask. The innovative LED array distribution on the inner aspect of an anatomically designed facial mask positively contributed to the consistency of clinical results owing to the almost constant minimal distance between light sources and facial skin surfaces. More studies are needed to identify maintenance treatment intervals to keep clinical results within an acceptable range, aiming at an effective, long-lasting 'healthy ageing' protocol.

► **Declaration of interest** Loan of equipment for research purposes

► **Figure images** © Dr Leonardo Marini

Key points

- Predominantly non-thermal high-energy LED photobiomodulation has proven effective in positively modifying skin tone and texture of prematurely aged facial skin
- Sequential combined narrow band CW + PW LED treatment was reported to be painless, safe and easy to perform
- The innovative LED array distribution on the inner aspect of an anatomically designed facial mask positively contributed to the consistency of clinical results

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