Die Wirksamkeit der Low Level Laser-Therapie (LLLT)

Viele klinische und wissentschaftliche Studien haben bewiesen, dass die Low Level Laser-Therapie für das Haarwachstum positive Resultate erzielen lässt. Einige dieser Studien haben wir für Sie aufgelistet.

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Low-level laser therapy as a treatment for androgenetic alopecia.

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Abstract

BACKGROUND AND OBJECTIVES:

Androgenetic alopecia (AGA) affects 50% of males by age 50 and 50% of females by age 80. Recently, the use of low-level laser therapy (LLLT) has been proposed as a treatment for hair loss and to stimulate hair regrowth in AGA. This paper aims to review the existing research studies to determine whether LLLT is an effective therapy for AGA based on objective measurements and patient satisfaction.

STUDY DESIGN:

A systematic literature review was done to identify articles on Medline, Google Scholar, and Embase that were published between January 1960 and November 2015. All search hits were screened by two reviewers and examined for relevant abstracts and titles. Articles were divided based on study design and assessed for risk of bias.

RESULTS:

Eleven studies were evaluated, which investigated a total of 680 patients, consisting of 444 males and 236 females. Nine out of 11 studies assessing hair count/hair density found statistically significant improvements in both males and females following LLLT treatment. Additionally, hair thickness and tensile strength significantly improved in two out of four studies. Patient satisfaction was investigated in five studies, and was overall positive, though not as profound as the objective outcomes.

CONCLUSION:

The majority of studies covered in this review found an overall improvement in hair regrowth, thickness, and patient satisfaction following LLLT therapy. Although we should be cautious when interpreting these findings, LLLT therapy seems to be a promising monotherapy for AGA and may serve as an effective alternative for individuals unwilling to use medical therapy or undergo surgical options. Lasers Surg. Med. © 2016 Wiley Periodicals, Inc.

The Growth of Human Scalp Hair in Females Using Visible Red Light Laser and LED Sources

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Background and Objectives: Low level laser (light) therapy (LLLT) has been demonstrated to promote hair growth in males. A double-blind randomized controlled trial was undertaken to define the safety and physiologic effects of LLLT on females with androgenic alopecia.

Methods: Forty-seven females (18–60 years old, Fitzpatrick I-IV, and Ludwig–Savin Baldness Scale I-2, I-3, I-4, II-1, II-2 baldness patterns) were recruited. A transition zone scalp site was selected; hairs were trimmed to 3 mm height; the area was tattooed and photographed. The active group received a "TOPHAT655" unit containing 21, 5 mW diode lasers (655 ± 5 nm) and 30 LEDS (655 ± 20 nm), in a bicycle-helmet like apparatus. The placebo group unit appeared identical, containing incandescent red lights. Patients treated at home every other day $\times$ 16 weeks (60 treatments, 67 J/cm^2 irradiance/25 minute treatment, 2.9 J dose), with follow up and photography at 16 weeks. A masked 2.85 cm^2 photographic area was evaluated by another blinded investigator. The primary endpoint was the percent increase in hair counts from baseline.

Results: Forty-two patients completed the study (24 active, 18 sham). No adverse events or side effects were reported. Baseline hair counts were 228.2 ± 133.4 ($N = 18$) in the sham and 209.6 ± 118.5 ($N = 24$) in the active group ($P = 0.642$). Post Treatment hair counts were 252.1 ± 143.3 (N = 18) in the sham group and 309.9 ± 166.6 ($N = 24$) in the active group ($P = 0.235$). The change in hair counts over baseline was 23.9 ± 30.1 ($N = 18$) in the sham group and 100.3 ± 53.4 ($N = 24$) in the active group ($P < 0.0001$). The percent hair increase over the duration of the study was 11.05 ± 48.30 ($N = 18$) for the sham group and 48.07 ± 17.61 ($N = 24$) for the active group ($P < 0.001$). This demonstrates a 37% increase in hair growth in the active treatment group as compared to the placebo group.

Conclusions: LLLT of the scalp at 655 nm significantly improved hair counts in women with androgenetic alopecia at a rate similar to that observed in males using the same parameters. Lasers Surg. Med. 46:601–607, 2014.

Key words: alopecia; clinical research; hair; human; laser; LED; low level laser therapy (LLLT); photobiomodulation; RCT

INTRODUCTION

Endre Mester first observed that mice treated with lasers during experiments investigating the potential carcinogenic effects of laser exposure regrew hair in shaved areas significantly faster than unexposed mice in 1967 [1,2]. Other investigators subsequently observed that

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some patients exhibited paradoxical hair growth at the periphery of areas treated with lasers for hair removal or adjacent to lesions treated with laser sources [3–5]. These seminal observations stimulated others to investigate the potential effects and applications of low level laser (light) therapy (LLLT) in male and female pattern androgenetic alopecia [6–15].

We have previously reported the results of the male arm of a randomized controlled trial that was undertaken to define the safety and physiologic effects that occur when the hair follicle and surrounding tissue structures of the human scalp are exposed to LLLT using a bicycle helmet type device fitted with an array of laser and LED light sources operating at 655 nm [16]. This laser system meets the requirements of an FDA Class 3R laser product, and as a non-medical laser system (RDW). The LED components are non-classified light sources when marketed for cosmetic applications, as is the case here. The device was granted an FDA 510k clearance for the treatment of males with Hamilton–Norwood IIa-V, or frontal patterns of hair loss, in patients with Fitzpatrick I-IV skin types based on the results for the male cohort of that trial [16,17].

The present investigation reports the results obtained for the female cohort of subjects treated under the TH655 study protocol.

MATERIALS AND METHODS

A clinical study was conducted as per the IRB approved TH655 protocol (Essex IRB, Lebanon, NJ). The trial was registered on www.ClinicalTrials.gov and was assigned the identifier NCT01437163. Forty-seven healthy female volunteers 18–60 years old were recruited at two IRB approved treatment sites.

Informed consent was obtained, and each female subject was screened to verify that she met the inclusion and exclusion criteria for the study. History and physical examinations were conducted. All 47 women had Fitzpatrick skin types I-IV and Ludwig–Savin Baldness Scale I-II (L-S I-2, I-3, I-4, II-1, II-2) baldness patterns. An area of scalp was selected in a transition zone at the vertex of the scalp at a site determined by the investigator. The hairs within the selected site were trimmed to a maximum height of 3 mm in area that was approximately 2.5 cm in diameter. The area was marked with a medical tattoo using green ink using aseptic technique.

The site was then photographed using a custom camera apparatus that consisted of a Canon Rebel T3i 18 Megapixel camera system (Canon USA, Melville, NY) equipped with a Tamron 60 mm f/2 Macro lens with 1:1 magnification (Tamron USA, Commack, NY). A 55 mm Lens attachment ring was used to affix a Promaster RL60 LED Ring Light (Promaster, Inc, Fairfield, CT). The camera system was mounted to a custom Stand-off device which was manually positioned onto the scalp surface by the investigator each time photographs were taken. Images were taken positioning the tattoo in the center of the frame. These baseline images were coded and then forwarded to the photographic consultant. The photographic consultant verified that the images were of acceptable quality and processed the images for transmission to the investigator responsible for conducting the hair counts. The transmitted images were masked using a black mask to produce a 1.9 cm diameter circle centered on the tattoo, which provided a consistent 2.85 cm^2 area for hair counts. Neither the photographic consultant nor the investigator performing the hair counts was aware of the identity of the subject or the subjects' study group assignment.

Subjects were randomly assigned to active treatment or placebo treatment groups. Each subject received a numbered "TOPHAT655" unit (Apira Science, Inc, Boca Raton, FL) which was distributed to her by the Project Manager, who also provided instructions for the care and use of the device. The patients, the treating physicians, the photographic consultant, and the investigator performing the hair counts were not aware whether the device was a therapeutic (active) device or a functioning placebo (sham). The investigational devices did not have corporate logos or other identifiers, with the exception of a study investigational device number. A serial number was assigned to each helmet, which was recorded in a device log that contained the reference code for placebo and actual test unit. This log was not revealed to any investigator, subject, office staff, hair counter or sponsor employee.

The active treatment group received a "TOPHAT 655" unit containing 21, 5 mW laser diodes and 30 LEDS both operating at 655 nm (655 ± 5 nm and 655 ± 20 nm, respectively) and providing constant illumination over the scalp under the apparatus. Each subject self-treated at home for 25 minutes per treatment session every other day for 16 weeks (60 treatments, 67 J/cm^2 delivered irradiance, and 2.9 J per treatment session).

The sham group received a unit that was identical in appearance and function to the laser group devices, with the exception that the light sources were incandescent wheat lights that were painted red to mimic the appearance and configuration of the functioning device. Each subject in the sham group self-treated at home for 25 minutes/treatment, every other day for 16 weeks (60 treatments). Incandescent sources were substituted 1:1 for each laser diode and LED source position on the sham helmet's interior.

The light output of the active treatment and sham treatment devices was determined using an Ophir Nova Display Power Meter equipped with a Model 30A-P-R-SH detector head (Ophir-Spiricon, LLC, Logan, UT). The active devices delivered an energy density of 67 J/cm^2 at 655 nm per 25 minute treatment session at the level of the scalp. The placebo units delivered no measurable light at scalp level. The active device design was such that constant illumination was delivered over the areas of the scalp covered by the device.

The operating temperatures of the active and placebo devices were matched and were measured using a Klein Tools Model IR 3000 Thermometer (Klein Tools, Lincolnshire, IL). The temperature of the units was $27.8 \pm 0.3^{\circ}C$ at the level of the electronics and $22.2 \pm 0.3^{\circ}C$ on the interior surface of the helmet.

Study treatments were self-administered as follows: The subject's head was self-positioned within the helmet, until a sensor triggered the start of therapy. There was no contact between the subject and the light-emitting device; only the light reaches the subject scalp. Treatment duration was set to 25 minutes. The lasers and LEDs automatically shut off after the treatment session was complete. All device function was controlled by a hand set that was actuated by the user subject once the power cord was plugged into a standard 120 volt outlet and the start button was pressed. All other functions were pre-programmed and automatic. A full set of user instructions accompanied each helmet. There was no pre or post treatment care required, only that subjects' hair must be clean and not contain spray or gel fixative agents. No safety eyewear was required during the treatment sessions. A complete demonstration of the proper use of the helmet was provided to each subject at the time the test units were distributed. Periodic subject monitoring was conducted by telephone. Subjects were queried relative to their use of the device and for any possible side effects or adverse events.

The subjects returned at 16 weeks for follow up and post treatment photography of the previously marked area. The area was again trimmed and photographed using the same apparatus and photographic conditions as at the initial (baseline) visit. The images were processed, transmitted and analyzed in the same fashion as was the case for the pretreatment photographs.

One pre-treatment (baseline) and one post-treatment image were counted for each subject. The number of terminal hairs present in the masked area was counted and recorded.

Data analysis was conducted by a consulting statistician, who was provided the raw data and who was blinded as to identify the subjects and their individual treatments. The primary endpoint for evaluation was the percent increase in hair counts from baseline at the end of 16 weeks of treatment. The percent increase from baseline is to be obtained by the following formula:

$$X = 100 \times \frac{\text{End Count} - \text{Baseline Count}}{\text{Baseline Count}}$$

A data pooling analysis was done to determine whether there was a site by treatment interaction in the percent increase. An analysis of variance was done with only site, treatment group, and site by treatment group interaction in the model and the interaction was not statistically significant. The data were pooled across both sites to arrive at an estimate of the effect for the primary endpoint. Univariate tests comparing the Sham and Active treatment groups were by Wilcoxon rank-sum tests, and an unequal variance t-test was performed.

RESULTS AND STATISTICAL ANALYSIS

Study Site Subject Distribution

The study was a blinded multicenter study. The study subjects were allocated to Active Treatment or Sham on a 1:1 basis at each of two study sites. The distribution of study subjects by random treatment assignment and study site are given in Table 1.

TABLE 1. Subjects, Treatment Assignments, and Study Sites

Site	Sham (Placebo)	Active Treatment	Total
1	6	7	13
2	12	17	29
Total	18	24	42

A total of 47 patients were enrolled in the study and completed baseline screening and photography. However, three subjects at site one and two subjects from site two withdrew from the study prior to the initiation of treatment. Thus there were 24 active treatment and 18 sham subjects available for analysis at the end of the study after 16 weeks of treatment.

There were no reported side effects or adverse events reported by any subject or site at any time during the conduct of the study.

Baseline Demographic Characteristics

There was information gathered on three important demographic characteristics, subject age, subject Fitzpatrick Skin Type, and Ludwig–Savin Baldness Scale. The results of these characteristics by treatment group are presented in the Table 2.

Note that age was not statistically significant by treatment group nor was it significant by study site ($P = 0.0320$). Neither Fitzpatrick skin type nor the Ludwig–Savin Baldness Scale differed by treatment group. Both study sites differed by Fitzpatrick Skin Type ($P < 0.001$) and by Ludwig–Savin Baldness Scale ($P < 0.001$).

Hair Counts and Photography

Photographs of the selected scalp site were taken prior to any treatment (baseline) and the same site was again photographed after the final treatment had been performed (post-treatment).

TABLE 2. Baseline Demographic Characteristics by Treatment Group

Characteristic	Sham (Placebo)	Active Treatment	P-value
Age			0.068
Mean (SD) N	51.00 (7.05) 18	46.29 (9.22) 24	
Med (Min, Max)	53 (33, 60)	49 (26, 58)	
Fitzpatrick Skin Type			0.582
I n (%)	3 (22.22)	4 (16.67)	
II n (%)	3 (16.67)	6 (25.00)	
III n (%)	12 (61.11)	12 (50.00)	
IV n (%)	0 (0.00)	2 (8.33)	
Ludwig-Savin Baldness Scale			0.858
I n (%)	7 (33.33)	11 (45.83)	
II n (%)	11 (66.67)	13 (54.17)	

Examples of baseline (pre treatment) and final (post treatment) images are presented in Figures 1 and 2. Figure 1 demonstrates the results for typical patients in the placebo or sham group. Note that there is only a slight change present in the images taken at 16 weeks as compared to the baseline images. Figure 2 demonstrates baseline and final images for typical subjects in the active treatment group. A significant increase in the number of terminal hairs present is evident in the 16 week photographs compared to baseline. The diameter of the hairs present in the sample areas was not measured.

Baseline Hair Counts

The analyses reported below were conducted in Minitab 16 (Minitab, Inc, State College, PA). The raw data for these analyses appear in Appendix 1.

The baseline hair counts by treatment group and study site are presented in Table 3. While the two study sites differ in the absolute values for the mean baseline hair counts, there was no statistical difference between the mean hair counts in the active and sham group subjects at the particular study center. An analysis of variance was done with only site, treatment group, and site by treatment group interaction in the model and the interaction was not statistically significant ($P = 0.812$).The study site was used as a possible covariate in the multivariable analyses performed below.

Primary Analysis

The primary endpoint was the percent increase in hair counts from baseline at the end of 16 weeks of treatment (60 treatments). The percent increase from baseline was obtained for each subject by using the formula above.

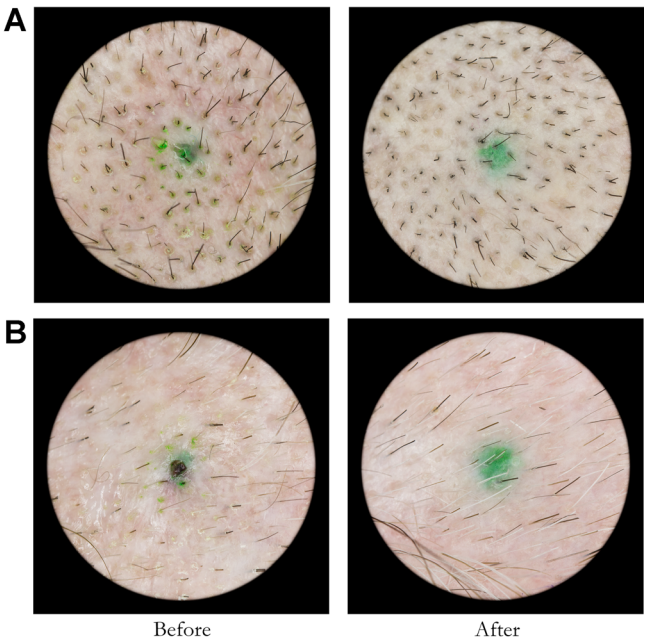

Fig. 1. Sham treatment group subject pre and post treatment image examples. Hair counts for subject A were 151 at baseline and 166 post treatment. Hair counts for subject B were 41 at baseline and 44 post treatment.

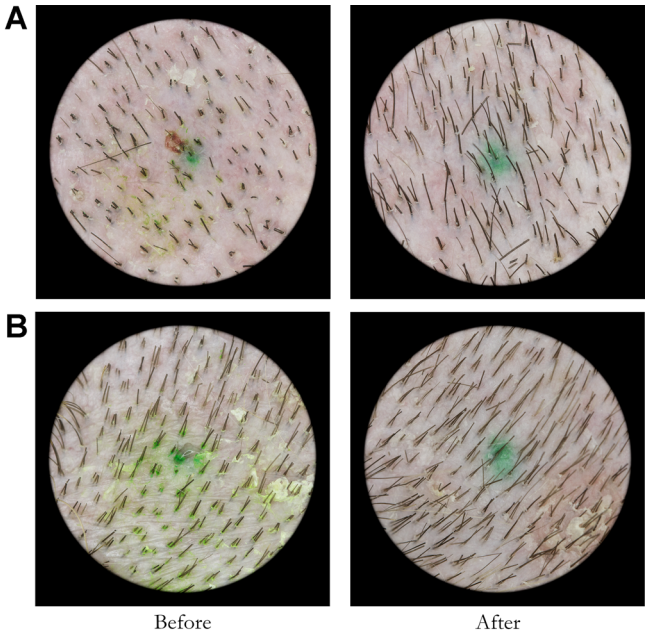

Fig. 2. Active treatment group subject pre and post treatment image examples. Hair counts for subject A were 153 at baseline and 221 post treatment. Hair counts for subject B were 108 at baseline and 209 post treatment.

A data pooling analysis was done to determine if there was a site by treatment interaction in the percent increase. If the interaction between site and treatment was significant with a $P < 0.15$, there would be evidence of a site by treatment interaction that would require weighting the site results to get an estimate of the study effect. An analysis of variance was done with only site, treatment group, and site by treatment group interaction in the model and the interaction was not statistically significant ($P = 0.812$). Thus the data were pooled across both sites to arrive at an estimate of the effect for the primary endpoint.

Univariate tests comparing the Sham and Active Treatment groups were intended to be by Wilcoxon rank-sum tests unless the variance between the two groups was statistically significantly different. In that case, the comparison was to be conducted by an unequal variance t-test. The results of the pooled data analysis appear in Table 4.

These results indicate that the univariate result comparing the increase in hair counts was statistically significant ($P = 0.001$). Low level laser treatment for 16 weeks increased mean hair counts by about 37% relative to sham treatment using the study device and the study treatment parameters. A multivariable analysis accounting for baseline differences in hair counts by study site indicates that the percent increase by treatment adjusted for study site indicate that the study site had a non-significant impact on the percent ($P = 0.218$). Therefore the

TABLE 3. Baseline Hair Counts of Vertex Scalp Site

Site	Sham (Placebo), Mean (SD) N, Med (Min, Max)	Active Treatment, Mean (SD) N, Med (Min, Max)	*P*-value
1	317.5 (174.1) 6, 277 (130, 560)	335.4 (144.6) 7, 260.0 (244, 599)	0.846[a]
2	183.5 (84.9) 12, 201.5 (41, 327)	157.8 (50.5) 17, 152.0 (53, 234)	0.361[a]
P-Value	0.125[a]	0.019[a]	—

[a]Two-sided unequal variance *t*-test.

study site differences in baseline counts did not modify the effect of treatment on the percent increase in hair counts after treatment. A second supportive multivariable analysis used baseline count as a covariate and in that analysis, the baseline term was not significant ($P = 0.627$), treatment was highly significant ($P < 0.0001$), but Study Site was not statistically significant ($P = 0.219$). Further, when age, Fitzpatrick type and Ludwig–Savin scale were included in a third sensitivity model, none were statistically significant with *P*-values of 0.901, 0.939, and 0.538, respectively. Thus, the univariate result is confirmed by the multivariable analysis with active LLLT treatment as the only significant term in the model ($P < 0.001$).

DISCUSSION

Treatment of androgenetic alopecia with LLLT has been studied in humans and in animal models using a variety of light sources, wavelengths and treatment parameters [6–9,11,12,14–16,18]. We previously reported the results of the TH655 RCT using the so-called TOPHAT 655 device in males with androgenetic alopecia [16].

The present study details the results of the female arm of the same study protocol, which was initiated and completed after the male study was concluded. These investigations employed a randomized, double-blind design and used a true placebo via a helmet identical in appearance to the active device, with incandescent sources that glowed red but did not deliver measurable light to the subject's scalp and which operated at a temperature of $22.2 \pm 0.3^{\circ}$C. Neither the active nor the sham devices delivered thermal energy to the scalp. Treatments were passive and did not depend on the user for delivery, aside from the subject being required to place the unit on the scalp and activate the controller.

Increases in hair counts were also observed in the sham or placebo group in the present study as was also the case in the earlier male cohort [16]. These observations may represent a true placebo effect, since the sham device did not deliver thermal energy or measurable light at scalp level. However, seasonal variations in hair growth or other factors could be the basis for this observation.

Avci et al. recently reviewed the use of LLLT for the treatment of hair loss [18]. They note that phototherapy is assumed to stimulate anagen re-entry in telogen hair follicles, prolong the duration of the anagen phase, increase the rates of proliferation in active anagen hair follicles and prevent premature catagen development [18]. They discuss several possible mechanisms for the photobiomodulation effect observed in these cases [18].

One such theory is that LLLT, particularly at wavelengths in the red range as was used in this investigation, affects the functioning of the stem cells that cause hair growth [16,18]. LLLT activates cytochrome c oxidase and increases mitochondrial electron transport [19–27], which leads to an increase in ATP and subsequent reversal of hair follicles from the dormant telogen stage of growth, to the active growth or anagen stage [6,7,9,11,13,14,16,18].

There is a growing body of evidence that the use of LLLT for the purpose of promoting hair growth is both safe and effective in both men and women. The optimal wavelengths and treatment parameters for treatment of alopecia remain indeterminate at this time. There is a need to conduct further studies in order to determine the potential role for near infrared and/or combinations of wavelengths as well as to investigate the effects of parameters such as coherence, pulsing and treatment frequency on clinical outcomes. The present study was not

TABLE 4. Baseline Hair Counts, End of Study Hair Counts, and Percent Increase by Treatment Group

Variable	Sham (Placebo), Mean (SD) N, Med (Min, Max)	Active Treatment, Mean (SD) N, Med (Min, Max)	*P*-value
Baseline	228.2 (133.4) 18, 216.5 (41, 560)	209.6 (118.5) 24, 187.5 (53, 599)	0.642[a]
Post Treatment	252.1 (143.3) 18, 248.0 (44, 636)	309.9 (166.6) 24, 270.5 (57, 829)	0.235[a]
Difference from Baseline	23.9 (30.1) 18, 15.5 (-23, 108)	100.3 (53.4) 24, 91.0 (4, 230)	<**0.0001**[a]
Percent Increase	11.05 (48.30) 18, 10.15 (-4.66, 43.20)	48.07 (17.61) 24, 45.58 (7.55, 93.52)	<**0.001**[a]

[a]Two-sided unequal variance *t*-test.

designed to investigate alternative treatment regimes or parameters. It was designed to evaluate the safety and effectiveness of a particular device designed for home use with specific parameters on the treatment of women with androgenetic alopecia.

We have demonstrated that the use of low level laser therapy at 655 nm applied to the scalp every other day for 16 weeks (60 treatments) via the TOPHAT 655 device resulted in a significant improvement in women who used the device. There was a 37% increase in terminal hair counts in the active treatment group as compared to the control (sham) group ($P < 0.001$) in 18–60 year old female subjects with I-2, I-3, I-4, II-1, or II-2 Ludwig–Savin baldness patterns and Fitzpatrick I-IV Skin Types. These results mirror those of the previously reported male trial which demonstrated a 35% increase in males with Hamilton–Norwood IIa-V baldness patterns and Type I–IV Fitzpatrick Skin Types [16].

Similarly, the female subjects were able to conduct the treatments at home and were able to apply and use the device as directed without any side effects or adverse events being reported at any time during the conduct of the study. This indicates that the device is safe for the unsupervised environment of home use and that the therapy is easily managed by both men and women using this device.

SUMMARY

The present study demonstrates that that low level laser (light) treatment of the scalp every other day for 16 weeks using the TOPHAT 655 device is a safe and effective treatment for androgenic alopecia in healthy women between the ages of 18–60 with Fitzpatrick Skin Types I–IV and Ludwig–Savin Baldness Scale I-2–II-2 baldness patterns. Subjects receiving LLLT at 655 nm achieved a 37% increase in hair counts as compared to sham treated control patients in this multicenter RCT. These results are similar to those reported in an earlier study using the same device in males with alopecia.

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APPENDIX A

Raw Hair Counts by Study Site and Treatment Group.

Subject[a]	Site	Treatment	Age (yrs)	Fitzpatrick Skin Type	Ludwig Savin Scale	Baseline Hair Count	Posttrt[b]	Diff[c]	Pct_bas[d]
1	1	Active	43	1	I	483	687	204	42.236
2*	1	—	27	1	II				
3	1	Sham	57	3	I	292	297	5	1.712
4*	1	—	45	1	I				
5	1	Sham	44	2	I	494	471	−23	−4.656
6	1	Active	52	1	I	245	333	88	35.918
7	1	Active	57	1	I	244	358	114	46.721
8*	1	—	49	3	I				
9	1	Sham	57	1	II	130	150	20	15.385
10	1	Active	50	1	II	249	334	85	34.137
11	1	Sham	33	1	I	560	636	76	13.571
12	1	Sham	58	3	II	262	311	49	18.702
13	1	Active	52	3	II	268	450	182	67.910
14	1	Active	52	2	I	260	354	94	36.154
15	1	Active	44	2	I	599	829	230	38.397
16	1	Sham	53	1	II	167	170	3	1.796
17	2	Active	44	3	I	228	375	147	64.474
18	2	Active	51	3	II	234	385	151	64.530
19	2	Active	50	3	II	145	221	76	52.414
20	2	Active	47	3	I	182	276	94	51.648
21	2	Active	33	3	II	153	221	68	44.444
22	2	Active	26	3	II	192	263	71	36.979
23	2	Active	56	3	II	148	203	55	37.162
24	2	Active	45	2	I	108	209	101	93.519
25	2	Active	44	3	II	53	57	4	7.547
26	2	Active	38	2	II	144	230	86	59.722
27	2	Active	51	3	II	152	265	113	74.342
28	2	Active	58	2	II	110	139	29	26.364
29	2	Active	53	3	II	225	340	115	51.111
30	2	Active	58	3	I	97	146	49	50.515
31	2	Sham	60	3	I	41	44	3	7.317
32	2	Sham	51	3	I	224	248	24	10.714
33	2	Sham	59	3	II	116	140	24	20.690
34	2	Sham	45	2	II	209	249	40	19.139
35	2	Sham	46	3	I	327	342	15	4.587
36	2	Sham	54	3	II	250	358	108	43.200
37	2	Sham	53	3	II	135	149	14	10.370
38	2	Sham	42	3	II	232	248	16	6.897
39*	2	—	20	3	I				
40	2	Sham	53	3	II	262	270	8	3.053
41	2	Sham	52	3	I	61	60	−1	−1.639
42	2	Active	28	4	I	204	328	124	60.784
43	2	Sham	55	2	II	151	166	15	9.934
44*	2	—	27	3	II				
45	2	Sham	46	3	II	194	229	35	18.041
46	2	Active	31	4	I	183	264	81	44.262
47	2	Active	48	2	II	124	171	47	37.903

[a]Patient numbers were grouped for convenience not by order of presentation or randomization.
[b]Posttrt is the hair count after 16 weeks of treatment.
[c]Diff = Posttrt – Baseline Hair Count.
[d]Pct_bas is the percent hair increase (decrease) at 16 weeks as a percent of baseline.
*Five subjects withdrew from the study after enrollment and prior to treatment.

The Growth of Human Scalp Hair Mediated by Visible Red Light Laser and LED Sources in Males

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Background and Objectives: Low level laser therapy (LLLT) has been used to promote hair growth. A double-blind randomized controlled trial was undertaken to define the safety and physiologic effects of LLLT on males with androgenic alopecia.

Methods: Forty-four males (18–48 yo, Fitzpatrick I–IV, Hamilton–Norwood IIa–V) were recruited. A transition zone scalp site was selected; hairs were trimmed to 3 mm height; the area was tattooed and photographed. The active group received a "TOPHAT655" unit containing 21, 5 mW lasers (655 ± 5 nm), and 30 LEDS (655 ± 20 nm), in a bicycle-helmet like apparatus. The placebo group unit appeared identical, containing incandescent red lights. Patients treated at home every other day $\times$ 16 weeks (60 treatments, 67.3 J/cm^2 irradiance/25 minute treatment), with follow up and photography at 16 weeks. A masked 2.85 cm^2 photographic area was evaluated by another blinded investigator. The primary endpoint was the percent increase in hair counts from baseline.

Results: Forty-one patients completed the study (22 active, 19 placebo). No adverse events or side effects were reported. Baseline hair counts were 162.7 ± 95.9 ($N = 22$) in placebo and 142.0 ± 73.0 ($N = 22$) and active groups respectively ($P = 0.426$). Post Treatment hair counts were 162.4 ± 62.5 ($N = 19$) and 228.7 ± 102.8 ($N = 22$), respectively ($P = 0.0161$). A 39% percent hair increase was demonstrated (28.4 ± 46.2 placebo, $N = 19$; 67.2 ± 33.4, active, $N = 22$) ($P = 0.001$) Deleting one placebo group subject with a very high baseline count and a very large decrease, resulted in baseline hair counts of 151.1 ± 81.0 ($N = 21$) and 142.0 ± 73.0 ($N = 22$), respectively ($P = 0.680$). Post treatment hair counts were 158.2 ± 61.5 ($N = 18$) and 228.7 ± 102.8 ($N = 22$) ($P = 0.011$), resulting in a 35% percent increase in hair growth (32.3 ± 44.2, placebo, $N = 18$; 67.2 ± 33.4, active, $N = 22$) ($P = 0.003$).

Conclusions: LLLT of the scalp at 655 nm significantly improved hair counts in males with androgenetic alopecia. Lasers Surg. Med. 45:487–495, 2013.

Key words: Alopecia; clinical research; hair; human, laser; LED; low level laser therapy (LLLT); photobiomodulation; RCT

INTRODUCTION

Low-level laser therapy (LLLT) has been studied and used for the treatment of a variety of clinical indications [1–5] including pain management [1,5], wound healing [2–21], and more recently to promote hair regrowth [22–36]. Each of these applications is based on

Conflict of Interest Disclosures: All authors have completed and submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest.

R.P. Chiacchierini and Eric Kazmirek have no disclosures. R.R. Blanche has received consulting fees, has had study related travel expenses paid and has ownership interest in Apira Science. A.B. Bodian has received equipment from Apira Sciences, has received discounts from Palomar, served on advisory boards for Allergan and Medicis, and speakers bureau for Palomar. A. Fernandez-Obregon has served on the speakers bureau for Amgen, Galderma, and Abbott. R.J. Lanzafame has received consulting fees from Apira Science and fees for manuscript preparation. He is Editor-in-Chief of Photomedicine and Laser Surgery, on the Editorial Boards of General Surgery News, Journal of Laparoendoscopic Surgery, Journal of the Society of Laparoscopic Surgeons, and Lasers in Medical Science. He serves as a consultant to the General and Plastic Surgery Devices and other panels of the Medical Devices Advisory Committee of the FDA's Center for Devices and Radiological Health. He performs medicolegal consulting for various law firms and entities. He serves as a consultant for various companies, including Business and venture capital groups including Leerink Swan, GLG Councils and others. He is member of the Board of Directors and Director of Continuing Medical Education for the American Society for Laser Medicine and Surgery. He is a partner in Biomedical Gateway, LLC, which was formed to seek grants in HIT, medical device development, and research.

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the principles of photobiomodulation which have demonstrated biological effects in living organisms [1–21].

The potential application of LLLT to stimulate hair growth can be traced to Endre Mester, a physician practicing in Budapest Hungary [22,23]. He discovered that mice treated with lasers during experiments designed to study the potential carcinogenic effects of laser exposure regrew the shaved hair in half the time of non-radiated mice. This 1967 study was the first reference to LLLT and hair growth. Other investigators noted the occurrence of paradoxical hair growth at the periphery of areas treated with lasers for hair removal or adjacent to lesions treated with laser sources [24–26].

These observations led to laboratory and clinical investigations on the effects and applications of LLLT in male and female pattern hair loss [27–36]. The HairMax LaserComb (Lexington International, LLC, Boca Raton, FL) is one such device that has been granted an FDA 510k clearance for use in treating males with Hamilton–Norwood IIa-V and females with Ludwig I-4, II-1, II-2, or frontal patterns of hair loss, in patients with Fitzpatrick I-IV skin types [32,35].

The present study aimed to define the safety and physiologic effects that occur when the human hair follicle and surrounding tissue structures are exposed to LLLT using a novel bicycle helmet type device that is fitted with an array of laser and LED light sources operating at 655 nm. This laser system is classified by the FDA as a class 3R laser, a non-medical laser system (RDW) and therefore, not subject to pre-market clearance or approvals. It may be marketed for hair wellness, which is defined as thicker, denser, more supple, and darker hair shafts. The LED components are non-classified light sources when marketed for cosmetic applications, as is the case here.

MATERIALS AND METHODS

A clinical study was conducted as per the IRB approved TH655 protocol (Essex IRB, Lebanon, NJ; Appendix 1). The trial is registered on www.ClinicalTrials.gov and is assigned the identifier NCT01437163. Forty-four healthy male volunteers 18–48 years old were recruited at two IRB approved treatment sites.

Informed consent was obtained, and the male patients were screened to verify that they met the inclusion and exclusion criteria for the study. History and physical examinations were conducted. All 44 patients had Fitzpatrick skin types I-IV and Hamilton–Norwood IIa-V baldness patterns. An area of scalp was selected in a transition zone at the vertex of the scalp at a site determined by the investigator and based on the individual patient's hair loss pattern. The hairs in the selected site were trimmed to a maximum height of 3 mm in area that was approximately 2.5 cm in diameter. The area was marked with a medical tattoo using green ink using aseptic technique.

The site was then photographed using a custom camera apparatus specifically configured for this purpose. The apparatus consisted of a Canon Rebel T3i 18Megapixel camera system (Canon USA, Melville, NY) equipped with a Tamron 60 mm f/2 Macro lens with 1:1 magnification (Tamron USA, Commack, NY). A 55 mm Lens attachment ring was used to affix a Promaster RL60 LED Ring Light (Promaster, Inc., Fairfield, CT). The camera system was then mounted to a custom Stand-off device which was then manually positioned onto the scalp surface by the investigator each time photographs were taken. Images were taken with the tattoo positioned in the center of the frame. These baseline images were coded and then forwarded to the photographic consultant. The photographic consultant verified that the images were of acceptable quality and processed the images for transmission to the investigator responsible for conducting the hair counts. The transmitted images were masked using a black mask to produce a 1.905 cm diameter circle centered on the tattoo, which provided a consistent 2.85 cm^2 area for hair counts. Neither the photographic consultant nor the investigator performing the hair counts was aware of the identity of the subject or the subjects' study group assignment.

Patients were randomly assigned to active treatment or placebo treatment groups. Each subject received a numbered "TOPHAT655" unit (Apira Science, Inc, Boca Raton, FL) which was distributed to him by the Project Manager, who also provided the patients with instructions for the care and use of the device. Neither the patients, the treating physicians at the clinical sites, the photographic consultant, nor the investigator performing the hair counts was aware whether the device was a therapeutic (active) or a functioning placebo (sham) device. The TOPHAT655 devices used in the study resembled a device currently marketed for home use. However, the investigational devices did not have any corporate logos or other identifiers with the exception of a study investigational device number. (Fig. 1A) serial number was assigned to each helmet, which was then recorded in a device log that contained the code for placebo and actual test unit reference. This log was not revealed to any investigator, subject, office staff, hair counter, or sponsor employee.

The active treatment group received a "TOPHAT655" unit containing 20, 5 mW lasers, and 31 LEDS both operating at 655 nm (655 ± 5 nm and 655 ± 20 nm, respectively) and providing constant illumination over the scalp under the apparatus (Fig. 1). Each subject self-treated at home for 25 minutes/treatment every other day for 16 weeks (60 treatments, 67.3 J/cm^2 delivered irradiance per treatment session).

The placebo or sham group received a unit that was identical in appearance and function to the laser group devices, with the exception that the light sources were incandescent wheat lights that were painted red to mimic the appearance and configuration of the functioning device. Each subject in the sham group self-treated at home for 25 minutes/treatment, every other day for 16 weeks (60 treatments). The interior view of the placebo device is shown in Figure 2. Note that incandescent sources were substituted 1:1 for each laser diode and LED source position on the helmet's interior.

The light output of the active treatment and sham treatment devices was determined using an Ophir Nova Display Power Meter equipped with a Model 30A-P-R-SH

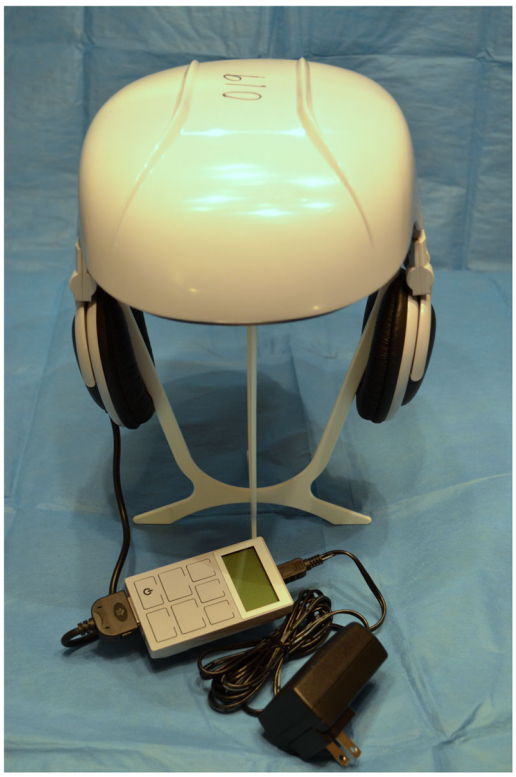

Fig. 1. The TOPHAT655 device unit exterior view. An example of the experimental device is shown with the control unit and power cord attached. Note that there are no identifying markings on the unit with the exception of the device number which is written on the top of the unit.

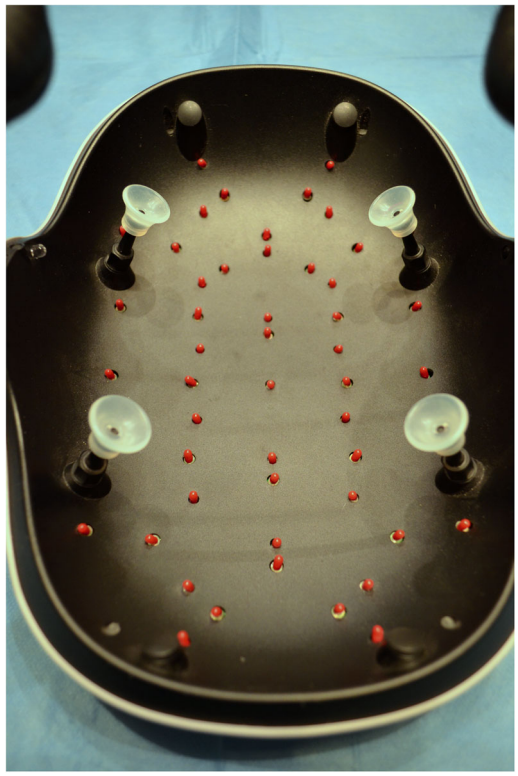

Fig. 2. The interior view of a placebo TOPHAT655 device unit. The interior view of a placebo unit is shown to illustrate the arrangement of the light sources within the unit. Incandescent panel lamps have been substituted for LED and Laser diodes at all light source locations on the helmet interior. Adjustable silicone bumpers allow for customized positioning on the subject's scalp.

detector head (Ophir-Spiricon, LLC, Logan, UT). The active devices delivered an energy density of 67.3 J/cm^2 at 655 nm per 25 minute treatment session at the level of the scalp. The placebo units delivered no measurable light at scalp level. The active device design was such that constant illumination was delivered over the areas of the scalp covered by the device.

The operating temperatures of the active and placebo devices were matched and were measured using a Klein Tools Model IR 3000 Thermometer (Klein Tools, Lincolnshire, IL). The temperature of the units was $27.78 \pm 0.3^{\circ}$C at the level of the electronics and $22.22 \pm 0.3^{\circ}$C on the interior surface of the helmet.

Study treatments were self-administered as follows: The subject's head was self-positioned within the helmet, until a sensor triggers the start of therapy. There was no contact between the subject and the light-emitting device; only the light reaches the subject scalp. Treatment duration was set to 25 minutes. The lasers and LEDs automatically shut off after the treatment session was complete. All device function was controlled by a hand set that was actuated by the user subject once the power cord was plugged into a standard 120 V outlet and the start button was pressed. All other functions were pre-programmed and automatic. A full set of user instructions accompanied each helmet. There was no pre or post treatment care required, only that subjects' hair must be clean and not contain spray or gel fixative agents. No safety eyewear was required during the treatment session. A complete demonstration of the proper use of the helmet was provided to each subject at the time the test units were distributed. Periodic subject monitoring was conducted by telephone. Subjects were queried relative to their use of the device and for any possible side effects or adverse events.

The subjects returned at 16 weeks for follow up and post treatment photography of the previously marked area. The area was again trimmed and photographed as per the initial visit. The photography was conducted using the same apparatus and conditions as at baseline. The images were processed, transmitted and analyzed in the same fashion as was the case for the pre-treatment photographs.

One pre-treatment (baseline) and one post-treatment image was counted for each subject. The number of terminal hairs present in the masked area was counted and recorded.

Data analysis was conducted by a consulting statistician, who was provided the raw data and who was blinded as to the identity of the subjects or their individual treatments. The primary endpoint for evaluation was the

percent increase in hair counts from baseline at the end of 16 weeks of treatment. The percent increase from baseline is the obtained by the following formula:

$$X = 100 \times \frac{(\text{End Count} - \text{Baseline Count})}{\text{Baseline Count}}$$

A data pooling analysis was done to determine whether there was a site by treatment interaction in the percent increase. An analysis of variance was done with only site, treatment group, and site by treatment group interaction in the model and the interaction was not statistically significant. The data were pooled across both sites to arrive at an estimate of the effect for the primary endpoint. Univariate tests comparing the Sham and Active treatment groups were by Wilcoxon rank-sum tests, and an unequal variance *t*-test was performed.

RESULTS AND STATISTICAL ANALYSIS

Study Site Subject Distribution

The study was a blinded multicenter study. The study subjects were allocated to Laser or Sham on a 1:1 basis at each of two study sites. The distribution of study subjects by random treatment assignment and study site are given in Table 1.

A total of 44 patients were enrolled in the study and completed baseline screening and photography. However, three subjects who were allocated to the sham group failed to return for 16-week evaluation at treatment site 2. Thus there were 22 patients in each group at baseline, but 22 laser and 19 sham patients were available for analysis at the end of the study after 16 weeks of treatment.

There were no reported side effects or adverse events reported by any subject or site at any time during the conduct of the study.

Hair Counts and Photography

Photographs of the selected scalp site were taken prior to any treatment (baseline) and the same site was again photographed after the final treatment had been performed (post-treatment).

Examples of baseline (pre-treatment) and final (post-treatment) images are presented in Figures 3 and 4. Figure 3 demonstrates the results for typical patients in the placebo or sham group. Note that there is minimal change in the 16-week study interval. Figure 4 demonstrates baseline and final images for typical subjects in the active treatment group. Note that there is a significant increase in the number of terminal hairs present and that the individual hairs subjectively appear to be thicker and more deeply pigmented than they were at baseline. However, the diameter of the hairs was not measured.

TABLE 1. Subjects, Treatment Assignments, and Study Sites

Site	Sham (placebo)	Active treatment	Total
1	13	13	26
2	9	9	18
Total	22	22	44

The distribution of study subject by treatment site and their assignments are shown.

Baseline Hair Counts

The analyses reported below were conducted in Minitab 16 (Minitab, Inc., State College, PA). The raw data for these analyses appear in Appendix 1.

The baseline hair counts by treatment group and study site are presented in Table 2. While the two study sites differ in the absolute values for the mean baseline hair counts, there was no statistical difference between the mean hair counts in the active and sham group subjects at the particular study center. An analysis of variance was done with only site, treatment group, and site by treatment group interaction in the model and the interaction was not statistically significant ($P = 0.094$). The study site was used as a possible covariate in the multivariable analyses performed below.

Primary Analysis

The primary endpoint was the percent increase in hair counts from baseline at the end of 16 weeks of treatment. The percent increase from baseline was obtained for each subject by using the formula above.

A data pooling analysis was done to determine if there was a site by treatment interaction in the percent increase. If the interaction between site and treatment was significant with a $P < 0.15$, there would be evidence of a site by treatment interaction that would require weighting the site results to get an estimate of the study effect. An analysis of variance was done with only site, treatment group, and site by treatment group interaction in the model and the interaction was not statistically significant ($P = 0.349$). Thus the data were pooled across both sites to arrive at an estimate of the effect for the primary endpoint.

Univariate tests comparing the Sham and Active Treatment groups were intended to be by Wilcoxon rank-sum tests unless the variance between the two groups was statistically significantly different. In that case, the comparison was conducted by an unequal variance *t*-test. The results of the pooled data analysis appear in Table 3.

These results indicate that the univariate result comparing the increase in hair counts was statistically significant ($P = 0.001$). The results indicate that low level laser treatment for 16 weeks increases mean hair counts by about 39%. A multivariable analysis accounting for baseline differences in hair counts by study site indicates that the percent increase by treatment adjusted for study site differences still had a significant effect ($P < 0.0001$). The study site differences in baseline counts did not diminish the effect of treatment on the percent increase in hair counts after treatment. A second supportive multivariable analysis used baseline count as a covariate and in that analysis, the baseline term was significant ($P = 0.035$),

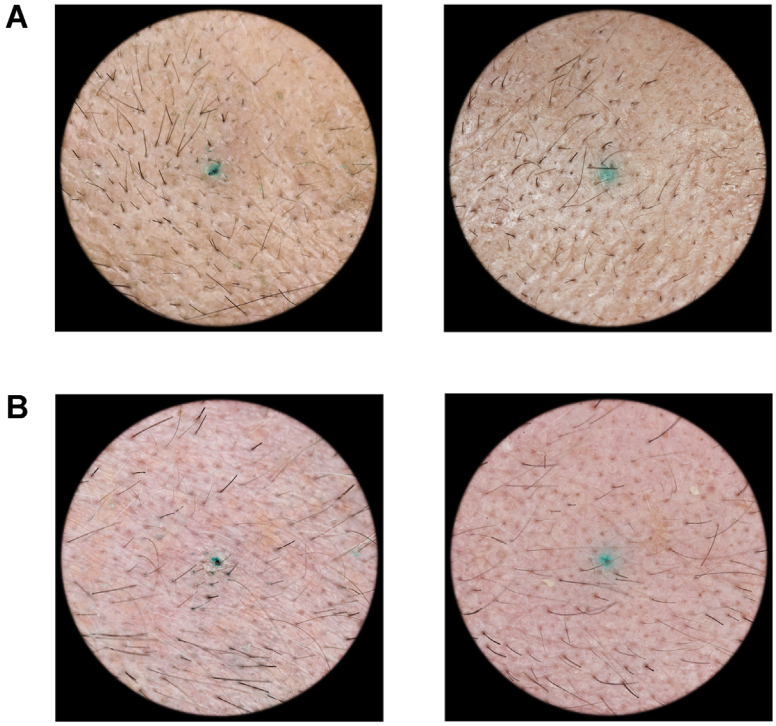

Fig. 3. Pre and post treatment image examples for Sham treatment group subjects. Pre-treatment and 16 weeks post-treatment photo pairs are shown for two placebo group subjects. Hair counts were 102 at baseline and 109 at 16 weeks in subject 83 (**A**) and 65 and 80, respectively in subject 93 (**B**).

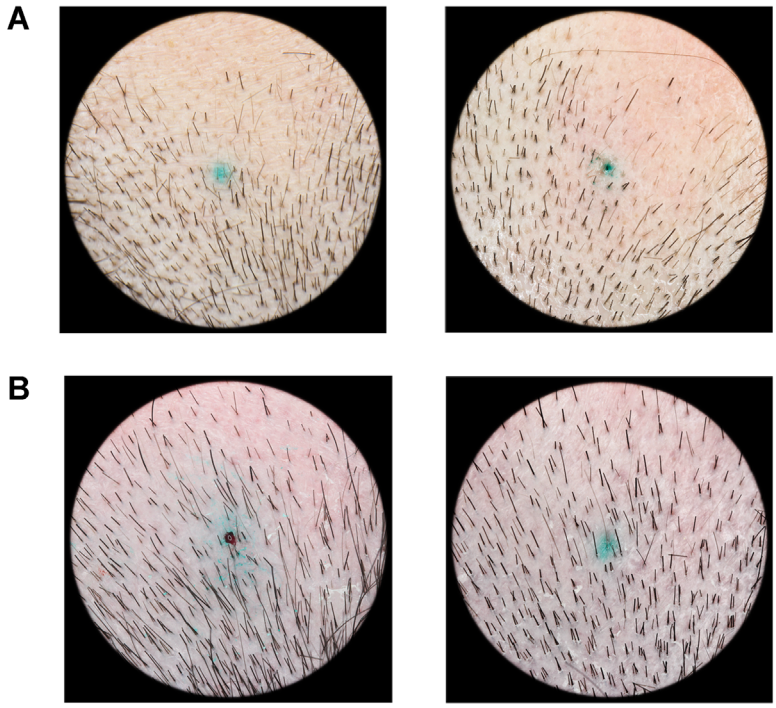

Fig. 4. Pre and post treatment image examples for active treatment group subjects. Pre-treatment and 16 weeks post-treatment photo pairs are shown for two active treatment group subjects. Hair counts were 140 at baseline and 280 at 16 weeks in subject 69 (**A**), and 143 and 322, respectively in subject 79 (**B**). Note that some of the hairs subjectively appear to be thicker and more deeply pigmented after treatment.

TABLE 2. Baseline Hair Counts of Vertex Scalp Site

Site	Sham mean (SD) N med (min, max)	Active treatment mean (SD) N med (min, max)	*P*-value
1	111.1 (49.7) 13 109 (29, 218)	101.0 (44.7) 13 97.0 (49, 205)	0.442[a]
2	237.3 (99.1) 9 334.5 (121, 406)	201.3 (65.4) 9 213.0 (81, 276)	0.691[a]
P-Value	0.005[b]	0.002[b]	—

The baseline hair count data is shown for the placebo and active treatment group patients and by treatment site at baseline. The mean ± SD, the median and range (min, max) are shown.
[a]Two-sided Wilcoxon rank sum test.
[b]Two-sided unequal variance *t*-test.

TABLE 3. Baseline Hair Counts, End of Study Hair Counts, and Percent Increase by Treatment Group

Variable	Sham mean (SD) N med (min, max)	Active treatment mean (SD) N med (min, max)	*P*-value
Baseline	162.7 (95.9) 22 134.0 (29, 406)	142.0 (73.0) 22 135.0 (49, 276)	0.426[a]
Post treatment	162.4 (62.5) 19 159.0 (63, 330)	228.7 (102.8) 22 237.5 (83, 403)	0.016[a]
Percent increase	28.4 (46.2) 19 12.4 (−41.4, 134.3)	67.2 (33.4) 22 59.2 (19.8, 127.3)	0.001[b]

The baseline hair count data is shown for the placebo and active treatment group patients and by treatment site after 16 weeks of therapy. The mean ± SD, the median and range (min, max) are shown.
[a]Two-sided unequal variance *t*-test.
[b]Two-sided Wilcoxon rank-sum test.

TABLE 4. Baseline Hair Counts, End of Study Hair Counts, and Percent Increase by Treatment Group Excluding Control Subject 3 at Site 2

Variable	Sham mean (SD) N med (min, max)	Active treatment mean (SD) N med (min, max)	*P*-value
Baseline	151.1 (81.0) 21 132.0 (29, 345)	142.0 (73.0) 22 135.0 (49, 276)	0.680[a]
Post Treatment	158.2 (61.5) 18 155.0 (63, 330)	228.7 (102.8) 22 237.5 (83, 403)	0.011[b]
Percent Increase	32.3 (44.2) 18 12.6 (−29.6, 134.3)	67.2 (33.4) 22 59.2 (19.8, 127.3)	0.003[a]

The baseline hair count data is shown for the placebo and active treatment group patients and by treatment site after 16 weeks of therapy. The mean ± SD, the median and range (min, max) are shown. Subject 3 from site 2 is excluded from this analysis as he had a high baseline hair count and a very large decrease relative to all other study subjects.
[a]Two-sided Wilcoxon rank-sum test.
[b]Two-sided unequal variance *t*-test.

treatment was highly significant ($P < 0.0001$), but Study Site was not statistically significant ($P = 0.094$). This analysis indicates that the baseline counts were the primary reason the study sites differed and adjusting for that effect reduces the significance of study site but does not affect the treatment difference.

It should be noted that one subject in the control group at Site 2 started with a very large baseline count and had a very large decrease. To see if this subject had an undue influence on the results, an analysis was done which deleted this subject from consideration. The test for Site by Treatment interaction for this analysis had $P = 0.527$ indicating the absence of an interaction. Thus the data were pooled and the analysis proceeded as above. The results of that analysis with the subject deleted from the pooled data are provided in Table 4.

These results indicate that the statistically significant increase in percent hair counts was not due to the single subject with a large decrease from baseline. The estimated mean percent increase deleting one subject was about 35%. Adjustment for differences in baseline counts by study site actually improved the statistical significance level and the result was minimally affected by removing one Sham subject with a very high loss after treatment.

DISCUSSION

Various investigators have studied a variety of light sources, wavelengths, and treatment parameters for the treatment of alopecia with LLLT [27–30,32,33,35,36]. Most of these reports on the efficacy of LLLT for alopecia have been prospective, uncontrolled, open label studies, and

have not been confirmed by multi-center, randomized, double blind, controlled trials (RCT) [27–30,33,35,36].

We have reported the results for an RCT of the so-called TOPHAT 655 device. The present study employed a randomized, double-blind design, and used a true placebo via a helmet identical in appearance to the active device, with incandescent sources that glowed red but did not deliver measurable light to the subject's scalp and which operated at a temperature of $22.22 \pm 0.3^{\circ}$C. Neither the active nor the sham devices delivered thermal energy to the scalp. Treatments were passive and did not depend on the user for delivery, aside from the subject placing the unit on the scalp, and activating the controller. This differs from the HairMax device studies that required the user to comb the scalp for a specified treatment time and employed a placebo device that was readily distinguished by the fact that it was a white light source [27–29,32,35].

Hair growth following exposure to low level laser therapy (LLLT) alone is not sufficient to document that photobiomodulation has occurred. Increases in hair counts were also observed in the sham or placebo group in the present study. These observations may represent a true placebo effect, since the sham device did not deliver thermal energy or measurable light at scalp level. However other explanations might also include seasonal variations in hair growth or other factors. This makes it important to include placebo and sham treatments in the study design and to conduct the investigation in such a manner as to minimize selection bias.

Several investigators have studied the effects of LLLT on hair growth in animal models [22,23,32,35]. Paradoxical hair growth after light based hair removal and other treatments in human subjects has also been observed with various laser and intense pulsed light sources [24–26,30].

The theory that is widely accepted is that LLLT, particularly at wavelengths in the red range as was used in this investigation, affects the functioning of the stem cells that cause hair growth. LLLT activates cytochrome c oxidase and increases mitochondrial electron transport [11–17], which leads to an increase in ATP and subsequent reversal of hair follicles from the dormant telogen stage of growth, to the active growth or anagen stage [27,28,30–32,34,35].

Analysis of non-radiated and radiated tissues has been employed to elucidate the tissue response and efficacy of the photobiomodulation effect [1,12–16,19–21]. However, the optimal wavelengths and treatment parameters remain indeterminate at this time. The present study was not designed to investigate alternative treatment regimes or parameters.

The ability of red light to stimulate hair follicle cellular proliferation and increase follicles in the anagen phase is supported by a preliminary study using the REVÅGE670 system (Apira Science, Boca Raton FL) [37]. This diode laser system operates at 670 nm and contains thirty 4 mW diode lasers affixed in a rotating helmet. Four subjects received two treatments per week for 6 weeks and one treatment per week for 6 weeks, totaling 18 laser treatments to the vertex of the scalp. Pretreatment and post treatment tissue samples were harvested after the 18th treatment. There were eight before and after biopsies taken from each subject. Four outcome measures were analyzed including: the number of hairs present, the presence of anagen hairs, the number of hairs containing Melanin, and the presence of Ki67 which is a marker of proliferating cells in the hair follicles. All of the subjects showed improvement in at least one of these measures on histological analysis [37].

The present study demonstrates that the use of LLLT at 655 nm as applied to the scalp on an every other day basis for 16 weeks (60 treatments) via the TOPHAT 655 device resulted in a significant improvement in patients who used the device. Specifically, there was a 35% increase in terminal hair counts in the laser group as compared to the control or sham treatment group ($P = 0.003$) in male patients who were 18–48 years of age and had IIa-V Hamilton–Norwood baldness patterns and were of Fitzpatrick Skin Types I-IV.

All of the patients in the study were able to apply and use the device as directed to self-administer their treatments at home. There were no side effects or adverse events reported by any of the study subjects at any time during the conduct of the study. This indicates that the device is safe for the unsupervised environment of home use.

SUMMARY

The present study demonstrates that that low level laser treatment of the scalp every other day for 16 weeks using the TOPHAT 655 device is a safe and effective treatment for androgenic alopecia in healthy males between the ages of 18–48 with Fitzpatrick Skin Types I-IV and Hamilton–Norwood IIa-V baldness patterns. Subjects receiving LLLT at 655 nm achieved a 35% increase in hair counts as compared to sham treated control patients in this multicenter RCT.

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APPENDIX 1. Raw Hair Counts by Study Site and Treatment Group

Patient[a]	Site	Treatment	BL[c]	Posttrt[d]	Diff[e]	Pct bas[f]
1	1	Active	49	99	50	102.0408
2	1	Active	102	161	59	57.84314
3	1	Active	134	280	146	108.9552
4	1	Active	72	111	39	54.16667
5	1	Active	97	141	44	45.36082
6	1	Active	97	196	99	102.0619
7	1	Active	66	150	84	127.2727
8	1	Active	58	116	58	100
9	1	Active	81	125	44	54.32099
10	1	Active	143	322	179	125.1748
11	1	Active	205	329	124	60.4878
12	1	Active	145	273	128	88.27586
13	1	Active	64	83	19	29.6875
14	1	Sham	99	159	60	60.60606
15	1	Sham	99	125	26	26.26263
16	1	Sham	109	123	14	12.84404
17	1	Sham	29	63	34	117.2414
18	1	Sham	112	127	15	13.39286
19	1	Sham	102	109	7	6.862745
20	1	Sham	169	190	21	12.42604
21	1	Sham	42	83	41	97.61905
22	1	Sham	70	164	94	134.2857
23	1	Sham	218	241	23	10.55046
24	1	Sham	136	151	15	11.02941
25	1	Sham	132	182	50	37.87879
26	1	Sham	127	198	71	55.90551
27	2	Active	221	340	119	53.84615
28	2	Active	213	343	130	61.03286
29	2	Active	253	324	71	28.06324
30	2	Active	136	227	91	66.91176
31	2	Active	275	339	64	23.27273
32	2	Active	167	324	157	94.01198
33	2	Active	81	97	16	19.75309
34	2	Active	276	403	127	46.01449
35	2	Active	190	248	58	30.52632
36	2	Sham	161	160	−1	−0.62112
37[b]	2	Sham	249			
38[b]	2	Sham	345			
39	2	Sham	406	238	168	−41.3793
40	2	Sham	192	196	4	2.083333
41	2	Sham	159	112	−47	−29.5597
42[b]	2	Sham	179			
43	2	Sham	324	330	6	1.851852
44	2	Sham	121	134	13	10.7438

[a]Patient numbers were grouped for convenience not by order of presentation or randomization.
[b]Three subjects refused to return for the 16 week assessment at site 2.
[c]BL is the baseline count.
[d]Psttrt is the hair count after 16 weeks of treatment.
[e]Diff = Psttrt–BL.
[f]Pct_bas is the percent hair increase (decrease) at 16 weeks as a percent of baseline.

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ORIGINAL RESEARCH ARTICLE

Efficacy and Safety of a Low-level Laser Device in the Treatment of Male and Female Pattern Hair Loss: A Multicenter, Randomized, Sham Device-controlled, Double-blind Study

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Abstract

Significance Male and female pattern hair loss are common, chronic dermatologic disorders with limited therapeutic options. In recent years, a number of commercial devices using low-level laser therapy have been promoted, but there have been little peer-reviewed data on their efficacy.

Objective To determine whether treatment with a low-level laser device, the US FDA-cleared HairMax Lasercomb®, increases terminal hair density in both men and women with pattern hair loss.

Methods Randomized, sham device-controlled, double-blind clinical trials were conducted at multiple institutional and private practices. A total of 146 male and 188 female subjects with pattern hair loss were screened. A total of 128 male and 141 female subjects were randomized to receive either a lasercomb (one of three models) or a sham device in concealed sealed packets, and were treated on the whole scalp three times a week for 26 weeks. Terminal hair density of the target area was evaluated at baseline and at 16- and 26-week follow-ups, and analyzed to determine whether the hypothesis formulated prior to data collection, that lasercomb treatment would increase terminal hair density, was correct. The site investigators and the subjects remained blinded to the type of device they dispensed/received throughout the study. The evaluator of masked digital photographs was blinded to which trial arm the subject belonged.

Results Seventy-eight, 63, 49, and 79 subjects were randomized in four trials of 9-beam lasercomb treatment in female subjects, 12-beam lasercomb treatment in female subjects, 7-beam lasercomb treatment in male subjects, and 9- and 12-beam lasercomb treatment in male subjects, compared with the sham device, respectively. Nineteen female and 25 male subjects were lost to follow-up. Among the remaining 122 female and 103 male subjects in the

Trial Registration: All trials were registered with http://www.clinicaltrials.gov. Trial #1 (registration #NCT00981461), "Treatment of Androgenetic Alopecia in Females, 9 Beam"; Trial #2 (#NCT01016964), "Treatment of Androgenetic Alopecia in Females, 12 Beam"; Trial #3 (#NCT00947505) and Trial #4 (#NCT00947219), "Treatment of Androgenetic Alopecia in Males".

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efficacy analysis, the mean terminal hair count at 26 weeks increased from baseline by 20.2, 20.6, 18.4, 20.9, and 25.7 per cm^2 in 9-beam lasercomb-treated female subjects, 12-beam lasercomb-treated female subjects, 7-beam lasercomb-treated male subjects, and 9- and 12-beam lasercomb-treated male subjects, respectively, compared with 2.8 ($p < 0.0001$), 3.0 ($p < 0.0001$), 1.6 ($p = 0.0017$), 9.4 ($p = 0.0249$), and 9.4 ($p = 0.0028$) in sham-treated subjects (95 % confidence interval). The increase in terminal hair density was independent of the age and sex of the subject and the lasercomb model. Additionally, a higher percentage of lasercomb-treated subjects reported overall improvement of hair loss condition and thickness and fullness of hair in self-assessment, compared with sham-treated subjects. No serious adverse events were reported in any subject receiving the lasercomb in any of the four trials.

Conclusions and relevance We observed a statistically significant difference in the increase in terminal hair density between lasercomb- and sham-treated subjects. No serious adverse events were reported. Our results suggest that low-level laser treatment may be an effective option to treat pattern hair loss in both men and women. Additional studies should be considered to determine the long-term effects of low-level laser treatment on hair growth and maintenance, and to optimize laser modality.

1 Introduction

Male and female pattern hair loss is a common, chronic dermatologic disorder. Male pattern hair loss (MPHL, or androgenetic alopecia, AGA) affects 50 % of men by 50 years of age, and the frequency and severity increase with age [1]. MPHL is characterized by a dihydrotestosterone-dependent process with miniaturization of terminal hair follicles (HFs) into vellus HFs [2]. The frequency and severity of female pattern hair loss (FPHL) also increase with age, with a prevalence of over 50 % in women over the age of 80 years [3]. While the role of androgens in all cases of FPHL is less certain, FPHL also undergoes follicular miniaturization [1]. Current medical treatments for pattern hair loss include topical minoxidil (available in 2 % and 5 % solutions or 5 % foam, and sometimes combined with other active ingredients such as tretinoin), finasteride, dutasteride (US FDA approved for the treatment of benign prostatic hyperplasia, and prescribed off-label for treatment of MPHL), topical ketoconazole, anti-androgens and estrogens (for FPHL), and follicular unit transplantation [4]. In addition, there are numerous oral supplements and topical treatments claimed to have hair growth-promoting or anti-hair loss effects that are marketed directly to the consumers, without independent data supporting the claims.

In recent years, low-level laser/light therapy (LLLT), or photobiomodulation or photobiostimulation, has been promoted to prevent hair loss and stimulate hair growth in both MPHL and FPHL. There have been a number of commercially available devices designed for home use (daily or several times a week), and they are relatively inexpensive compared with current medical treatment and hair transplantation surgery. However, there have been few peer-reviewed data on efficacy [5]. In one published study, only seven subjects with pattern hair loss (six female subjects and one male subject) were evaluated upon treatment with a laser "hood" [6]. The study was not sham device-controlled and the results did not reach statistical significance. A more recent, randomized, double-blind, sham device-controlled trial found "TOPHAT655" (a helmet-like device with lasers and light-emitting diodes) treatment to increase terminal hair count in pattern hair loss, but only male subjects were included in the trial [7]. To date, the most comprehensive published study is a randomized, double-blind, sham device-controlled clinical trial of 110 male subjects showing that the HairMax Lasercomb® (Lexington International, LLC., Boca Raton, FL, USA), FDA-cleared to treat pattern hair loss in male subjects at the time, was effective in increasing terminal hair density after 26 weeks of treatment [8]. The device has since been approved for treating FPHL, though there has been only one published study supporting the efficacy, with limitations [9]. In this study, only seven female subjects were included. They were given a lasercomb to use for 6 months, and the terminal hair count was compared between baseline and at the end of the study. The FDA considered the LaserComb® a medical device of "moderate risk", therefore it only screened for safety, not efficacy.

Given the prevalence of MPHL and FPHL, their limited medical treatment and the high costs of hair transplantation, and the ready availability and user friendliness of LLLT home devices, it is important to determine whether LLLT can provide an effective alternative for pattern hair loss, especially FPHL, for which no randomized, controlled trials have been published. The objective of this study was to determine the efficacy of LaserComb® treatment of pattern hair loss in both male and female subjects, in four randomized, multicenter, sham device-controlled, double-blind prospective trials. A total of 122 female and 103 male subjects were included in the efficacy analysis after 26 weeks of treatment, and three lasercomb models were evaluated.

2 Methods

2.1 Patient Enrollment

The study protocol was evaluated under Good Clinical Practice guidelines and approved by the authors'

Institutional Review Boards (IRBs) or the Chesapeake Research Review, Inc. All trials were registered with http://www.clinicaltrials.gov. Prior to participation in the trials, each subject provided a written informed consent. Participants received free evaluations at baseline and at follow-ups. They were compensated for each visit and were given a lasercomb at the end of the study (26-week visit). Subject screening, recruitment, and follow-up were carried out at multiple study sites: Trial #1 (registration #NCT00981461), "Treatment of Androgenetic Alopecia in Females, 9 Beam": International Dermatology Research, Inc. (Miami, FL, USA), The Education & Research Foundation, Inc. (Lynchburg, VA, USA); Trial #2 (#NCT01016964), "Treatment of Androgenetic Alopecia in Females, 12 Beam": The Cleveland Clinic Foundation (Cleveland, OH, USA), University of Minnesota (Minneapolis, MN, USA), University of Miami Miller School of Medicine (Miami, FL, USA); Trial #3 (#NCT00947505) and Trial #4 (#NCT00947219), "Treatment of Androgenetic Alopecia in Males": Dermatology Consulting Services (High Point, NC, USA); Trials #1, #3, and #4: DermResearch, Inc. (Austin, TX, USA), Skin Laser and Surgery Specialist (Hillsborough, NJ, USA), and Palm Beach Research Center (West Palm Beach, FL, USA). Full trial protocol is available upon request.

2.1.1 Study Inclusion/Exclusion Criteria

To be included in the trials, subjects must have been healthy, 25–60 years of age, with active androgenetic hair loss (Norwood–Hamilton classification of IIa–V for male subjects [10] and Ludwig/Savin classification of I-4, II-1, II-2, or frontal for female subjects) [11–13] and have Fitzpatrick skin type I–IV [14]. Race/ethnicity information was collected. Subjects must not have taken or used the following medications within 6 months prior to screening: minoxidil, finasteride (or any other 5α-reductase inhibitors), medications with anti-androgenic properties (e.g., cyproterone acetate, spironolactone, ketoconazole, flutamide, bicalutamide), topical estrogen, progesterone, tamoxifen, anabolic steroids, medication that can potentially cause hypertrichosis (e.g., cyclosporine, diazoxide, phenytoin, psoralens), oral glucocorticoids (inhaled glucocorticoids were permitted), lithium, phenothiazines, or other medications at the discretion of the investigators. Other excluded medications were phytotherapy (e.g., saw palmetto) within 8 weeks, isotretinoin within the past year, and anticoagulation use [other than aspirin (<325 mg every day, which was stable for 3 months)]. Subjects were excluded if they had malignancy in the target area within 5 years, active infection on the scalp, chronic dermatologic conditions (e.g., eczema, psoriasis, infection) of the scalp other than pattern hair loss, a history of poor wound healing or keloid formation, a history of thyroid or other medical condition that might influence hair growth and loss; human immunodeficiency virus infection, possession of a pacemaker, defibrillator, or other active implantable device; a history of drug and/or alcohol abuse within the past 12 months; or any other medical conditions at the discretion of the investigators. Pregnant female subjects or female subjects planning on becoming pregnant during the duration of the study were excluded. Subjects with a history of photosensitivity to laser light, hair transplantation, scalp reduction, radiation to the scalp or chemotherapy within the past year, current hair weave or tattooing, as well as subjects with hair shorter than one-half inch or with light-blonde hair were also excluded.

2.2 The Lasercomb and Sham Devices

Three different lasercomb configurations were evaluated for similar laser dose rates. These models were designed to meet varying marketing demands, and the FDA required clinical studies on each model to ensure consistency of results. The 7- and 9-beam lasercombs (HairMax LaserComb®, Lexington International, LLC) emit 7 or 9 red laser beams (beam diameter <5 mm) at a wavelength of 655 nm (±5 %). The 12-beam dual model emits 6 beams at a wavelength of 635 nm (±5 %) and 6 beams at 655 nm (±5 %). The lasers for each device were identical in power output, and the treatment time was adjusted for similar laser dose rates: 15 min for the 7-beam model, 11 min for the 9-beam model, and 8 min for the 12-beam model. Two sham devices that emitted white light from light-emitting diode bulbs had identical appearance as the 7- and 9-beam lasercombs, and were used as controls for the 7-, and 9- or 12-beam lasercombs, respectively.

2.3 Study Design

Four multicenter prospective trials were designed, to be randomized, sham controlled, and double blind. In Trials #1 and #2, subjects with FPHL used a 9-beam (#1) or a dual 12-beam (#2) lasercomb and sham device. In Trials #3 and #4, subjects with MPHL used a 7- (#3) or a 9- or 12-beam (#4) lasercomb and sham device.

Each study protocol was approved by institutional or the Chesapeake IRB. Each Clinical Study Sponsor confirmed performance in compliance with Good Clinical Practice (GCP, as defined in CPMP/ICH/135/95), the Declaration of Helsinki (with amendments), and local legal and regulatory requirements. Lexington International LLC, as a company, is and has been compliant and certified to ISO9001 and ISO13485 Quality Standards. Lexington's Clinical Study Practices have been audited by the FDA and have confirmed to be in compliance with the FDA's GCP. All

studies were managed and audited by Palm Beach CRO (Clinical Research Organization) and validated to be in compliance with the approved protocol.

For subjects who met the inclusion and exclusion criteria, at the baseline visit, a "target site" in the affected scalp area was chosen using a 25 mm × 25 mm plastic template, and hair within this target site (25mm × 25mm) was clipped. The target site was then marked with a semi-permanent tattoo using a professional tattooing machine (K.P. Permanent Make-Up, Inc., Pomona, CA, USA), and photographed.

Each subject was then provided with either a lasercomb or a sham device. Randomization was generated by Eugene R. Heyman (http://www.erhstats.com) using the SAS PROC RAND method. For the 9- and 12-beam trial in male subjects (#4), randomization was generated 1:1:1 with a block size of 3. For all other trials, randomization was 2:1 with a block size of 3. The lasercomb and sham devices, along with instructions, were provided to the site investigators in sealed, sequentially numbered opaque packets in a blinded manner, and were dispensed sequentially. Both the site investigators and the subjects remained blinded to the type of device they dispensed/received throughout the study.

The subjects were instructed to apply the device three times per week, with the beam on, to their entire scalp; the duration of treatment specific for each device and their respective sham control was included in the sealed packet (15 min for the 7-beam model, 11 min for the 9-beam model, and 8 min for the 12-beam model). Each subject was required to keep a diary of usage, which was reviewed by the site investigator at the time of office visits. The study duration was 26 weeks, with clinical monitoring visits at 8, 16, and 26 weeks. Dermatology scalp assessment, safety assessment, global and macro digital imaging after hair clipping, and computer-aided hair counts of the target sites were performed by blinded investigators at weeks 16 and 26, and compared with baseline.

2.3.1 *Efficacy Evaluation*

Change of terminal hair density (hair count/cm^2) at 26 weeks from baseline was used as the endpoint to evaluate the efficacy of lasercomb treatment in male and female subjects with pattern hair loss. The Canfield Epilume System was used for digital imaging of the target sites at baseline and at weeks 16 and 26. All macro photographs, with a 10-mm scale bar divided in 0.1-mm increments, were labeled only by subject number and uploaded to an online database. An independent evaluator not connected to the clinical trials analyzed the uploaded images and performed computer-assisted hair counts, using the TrichoScience software (Tricholog, Moscow, Russia). The evaluator was a hair transplant surgeon with 20 years of experience in evaluation of hair counts, and was blinded to which trial arm the subject belonged, as well as which images were from baseline and which were from follow-up. Subjects also filled out questionnaires for self-assessment of overall improvement of hair loss condition and thickness and fullness of hair at the 16- and 26-week visits.

2.4 Statistical Analysis

Based on previous testing data on lasercomb use, change in terminal hair count from baseline to study endpoint was found to be a mean increase of just under 30 hairs/cm^2 with a standard deviation of 18.6 hairs/cm^2. For the sample size calculation, the assumed standard deviation was 20 hairs/cm^2 and the treatment difference was assumed to be 17 hairs/cm^2. Each trial had a planned enrollment of 60 subjects in a 2:1 allocation of lasercomb:sham device to achieve at least 80 % power while allowing a 10 % drop-out rate. In Trials #1–3, subjects were randomized to a 2:1 allocation of the lasercomb:sham device. In Trial #4, subjects were randomized in a 1:1:1 allocation of the 9-beam:12-beam:sham device. For subject enrollment, continuous variables (e.g., age) were analyzed with a one-way analysis of variance and categorical variables with the Fisher's exact test.

The primary efficacy endpoint was the change in terminal hair density within the target area at 26 weeks from baseline, assessed in all subjects with baseline and at least one post-randomization efficacy evaluation. The lasercomb-treated group was compared with the sham device group using least squares mean with two-sided at a 5 % level of significance. The primary analysis of efficacy was an analysis of co-variance, which modeled terminal hair density as a function of treatment group, study center, age (as a continuous variable), and Fitzpatrick skin type (as a categorical variable). The secondary efficacy endpoint was the categorical change in terminal hair density from baseline, analyzed using the Cochran–Mantel–Haenszel row mean score test with integer scores stratified by study site. Cochran's Q test was performed to analyze the homogeneity of results across genders, all trials, and all lasercomb models. Subject self-assessment was also evaluated using the Cochran–Mantel–Haenszel row mean score test with integer scores stratified by site. The DerSimonian–Laird approach was used to perform the meta-analysis homogeneity assessment. All statistical analyses were contracted to Stat-Tech Services, LLC (Chapel Hill, NC, USA). For evaluation of safety, adverse events were summarized and each event was evaluated for frequency.

3 Results

3.1 Study Population

A total of 188 female and 146 male subjects were screened, and 141 female and 128 male subjects were randomized to receive the lasercomb or sham device. Of these subjects, 19 female and 25 male subjects were lost to follow-up, leaving 122 female and 103 male subjects completing at least one follow-up. Sixty-five and 57 subjects (122 total) were included in the efficacy evaluation for Trials #1 and #2 (the female trials evaluating the 9-beam and dual 12-beam lasercomb, respectively) (Fig. 1; Table 1), and 38 and 65 subjects (103 total) were included in the efficacy evaluation for Trials #3 and #4 (the male trials evaluating the 7- and the 9- or 12-beam lasercomb, respectively) (Fig. 1; Table 2). There were no statistically significant differences in demographic characteristics or hair loss features between the lasercomb and sham group in any of the four trials at baseline (Tables 1 and 2). The age of the subjects was 25–61 years, and 94.7 % were Caucasian. The last follow-up was conducted after 26 weeks of treatment, an accepted standard for clinical trials on hair growth.

3.2 Analysis of Efficacy

The trials were designed to be randomized and double blind. Data from different study sites were pooled for statistical analysis. All the randomized subjects who had a baseline and at least one post-randomization evaluation were included in the efficacy analysis (Fig. 1). To account for dropouts thereafter, all data are presented in last observation carried forward for the analysis of covariance for Trials #1 and #4.

3.2.1 Primary Efficacy Analysis

In Trial #1, a significant difference in terminal hair density change from baseline was observed between the 9-beam lasercomb- and sham-treated female subjects at 26 weeks ($p < 0.0001$) (Fig. 2a). The lasercomb-treated subjects showed a much higher increase in terminal hair density compared with sham-treated subjects, with a mean of 20.2 ($\pm$11.2 standard deviation [SD]) versus 2.8 ($\pm$16.5 SD) per cm^2 (Fig. 2a). Similar improvement in terminal hair density was observed with the 12-beam lasercomb treatment in Trial #2 (Fig. 2b). The lasercomb-treated female subjects

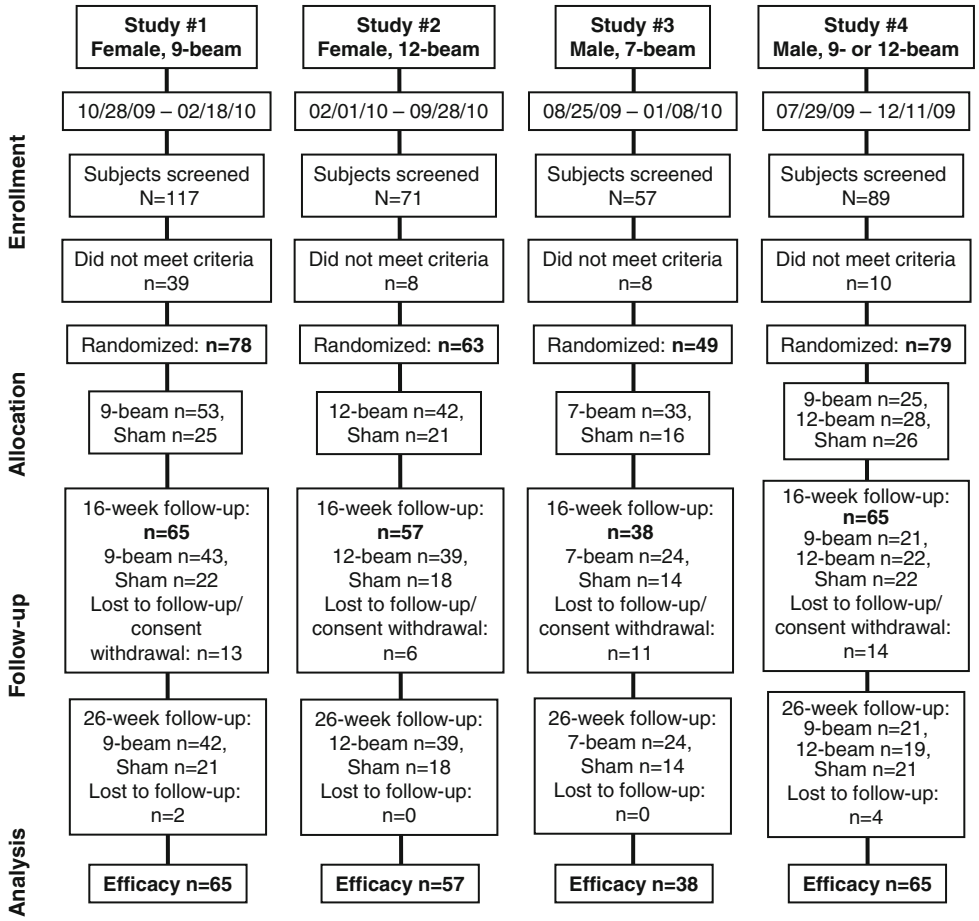

Fig. 1 Profile of the four randomized, sham-controlled trials of lasercomb treatment of male and female pattern hair loss. Dates of recruitments are indicated

Table 1 Demographic characteristics of female subjects at baseline for the 9- and 12-beam lasercomb trials

	Trial #1 ($n = 65$)			Trial #2 ($n = 57$)		
	9-beam lasercomb	Sham	p value	12-beam lasercomb	Sham	p value
Number of subjects	43	22		39	18	
Age (years)			0.8261			0.9102
Mean age (SD)	49.3 (9.1)	49.8 (7.3)		48.7 (10.2)	49.1 (8.3)	
Median age	52	49		50	49	
Range	29–60	37–60		26–61	33–60	
Race, *n* (%)			1.0000			1.0000
Caucasian	39 (90.7 %)	20 (90.9 %)		37 (94.9 %)	18 (100.0 %)	
African American	1 (2.3 %)	0 (0 %)		1 (2.6 %)	0 (0 %)	
Native American	0 (0 %)	0 (0 %)		0 (0 %)	0 (0 %)	
Alaska Native	0 (0 %)	0 (0 %)		0 (0 %)	0 (0 %)	
Asia/Pacific Islander	2 (4.7 %)	1 (4.5 %)		1 (2.6 %)	0 (0 %)	
Other	1 (2.3 %)	1 (4.5 %)		0 (0 %)	0 (0 %)	
Ethnicity, *n* (%)			0.2773			1.0000
Hispanic or Latino	13 (30.2 %)	10 (45.5 %)		10 (25.6 %)	4 (22.2 %)	
Not Hispanic or Latino	30 (69.8 %)	12 (54.5 %)		29 (74.4 %)	14 (77.8 %)	
Ludwig/Savin classification, *n* (%)			0.6513			0.2926
I-4	12 (27.9 %)	3 (13.6 %)		21 (53.8 %)	6 (33.3 %)	
II-1	11 (25.6 %)	7 (31.8 %)		11 (28.2 %)	6 (33.3 %)	
II-2	15 (34.9 %)	9 (40.9 %)		6 (15.4 %)	4 (22.2 %)	
Frontal	5 (11.6 %)	3 (13.6 %)		1 (2.6 %)	2 (11.1 %)	
Fitzpatrick skin type, *n* (%)			1.0000			0.7606
I	0 (0 %)	0 (0 %)		2 (5.1 %)	0 (0 %)	
II	15 (34.9 %)	7 (31.8 %)		11 (28.2 %)	4 (22.2 %)	
III	20 (46.5 %)	11 (50.0 %)		14 (35.9 %)	9 (50.0 %)	
IV	8 (18.6 %)	4 (18.2 %)		12 (30.8 %)	5 (27.8 %)	
Mean baseline hair count[a] (SD)	162.6 (46.2)	155.7 (43.5)		142.2 (40.5)	168.4 (41.1)	

[a] Number of terminal hairs per cm^2 in the target area

SD standard deviation

had a mean increase in terminal hair density of 20.6 (±11.6 SD) compared with 3.0 (±9.3 SD) for the sham group (Fig. 2b). Overall, primary efficacy analysis showed the difference in terminal hair density change at 26 weeks from baseline between lasercomb and sham treatment was highly significant ($p < 0.0001$) in both female trials (Fig. 2a, b). Similarly, statistically significant improvement was observed with lasercomb treatment compared with sham treatment in both male trials (Trial #3, 7-beam lasercomb vs. sham, $p = 0.0017$, Fig. 2c; Trial #4, 9- and 12-beam lasercombs vs. sham, $p = 0.0249$ and $p = 0.0028$, for the 9- and 12-beam lasercombs, respectively, Fig. 2d).

3.2.2 *Secondary Efficacy Analyses*

Secondary efficacy analyses included categorical summaries and covariate analyses of changes in terminal hair density from baseline. In Trial #1, 41 of 43 (95 %) of the 9-beam lasercomb-treated female subjects had hair density improvement of >5 hairs/cm^2 at 26 weeks while only 7 of 22 (32 %) sham-treated female subjects did ($p < 0.0001$) (Fig. 2e). Additionally, none of the 43 lasercomb-treated subjects showed decreased hair density as opposed to 11 of 22 (50 %) sham-treated subjects (Fig. 2e). Analysis of data collected at 16 weeks revealed similar results (data not shown). In Trial #2, 37 of 39 (95 %) of the 12-beam lasercomb-treated female subjects had hair density improvement of >5 hairs/cm^2 while only 6 of 18 (33 %) sham-treated female subjects did ($p < 0.0001$) (Fig. 2f). Although 7 of 18 (39 %) sham-treated subjects showed decreased hair density, only 1 of 39 (3 %) lasercomb-treated subjects did (Fig. 2f).

In Trial #3, 20 of 24 (83 %) of the 7-beam lasercomb-treated male subjects had hair density improvement of >5 hairs/cm^2, while only 6 of 14 (43 %) sham-treated male subjects did ($p = 0.0033$) (Fig. 2g). Additionally, only 2 of the 24 (8 %) lasercomb-treated male subjects showed

Table 2 Demographic characteristics of male subjects at baseline for the 7-, 9-, or 12-beam lasercomb trials

	Trial #3 ($n = 38$)			Trial #4 ($n = 65$)			
	7-beam lasercomb	Sham	p value	9-beam lasercomb	12-beam lasercomb	Sham	p value
Number of subjects	24	14		21	22	22	
Age (years)			0.0327				0.7100
Mean age (SD)	47.8 (9.0)	40.9 (9.5)		45.6 (9.3)	47.9 (9.6)	45.9 (10.4)	
Median age	48	41.5		50	50.5	47	
Range	26–59	25–55		26–58	26–59	30–61	
Race, n (%)			1.0000				1.0000
Caucasian	23 (95.8 %)	13 (92.9 %)		21 (100.0 %)	21 (95.5 %)	21 (95.5 %)	
African American	0 (0 %)	0 (0 %)		0 (0 %)	0 (0 %)	0 (0 %)	
Native American	0 (0 %)	0 (0 %)		0 (0 %)	0 (0 %)	0 (0 %)	
Alaska native	0 (0 %)	0 (0 %)		0 (0 %)	0 (0 %)	0 (0 %)	
Asia/Pacific islander	0 (0 %)	0 (0 %)		0 (0 %)	0 (0 %)	1 (4.5 %)	
Other	1 (4.2 %)	1 (7.1 %)		0 (0 %)	1 (4.5 %)	0 (0 %)	
Ethnicity, n (%)			0.6497				0.041
Hispanic or Latino	3 (12.5 %)	3 (21.4 %)		4 (19.0 %)	1 (4.5 %)	0 (0 %)	
Not Hispanic or Latino	21 (87.5 %)	11 (78.6 %)		17 (81.0 %)	21 (95.5 %)	22 (100.0 %)	
Norwood–Hamilton classification, n (%)			0.9130				1.0000
II	0 (0 %)	0 (0 %)		0 (0 %)	0 (0 %)	1 (4.5 %)	
III	10 (41.7 %)	5 (35.7 %)		10 (47.6 %)	10 (45.5 %)	10 (45.5 %)	
IV	9 (37.5 %)	5 (35.7 %)		8 (38.1 %)	9 (40.9 %)	7 (31.8 %)	
V	5 (20.8 %)	4 (28.6 %)		3 (14.3 %)	3 (13.6 %)	4 (18.2 %)	
Fitzpatrick skin type (%)			0.7904				0.998
I	1 (4.2 %)	0 (0 %)		3 (14.3 %)	2 (9.1 %)	2 (9.1 %)	
II	3 (12.5 %)	3 (21.4 %)		9 (42.9 %)	10 (45.5 %)	9 (40.9 %)	
III	12 (50.0 %)	5 (35.7 %)		7 (33.3 %)	8 (36.4 %)	9 (40.9 %)	
IV	8 (33.3 %)	6 (42.9 %)		2 (9.5 %)	2 (9.1 %)	2 (9.1 %)	
Mean baseline hair count[a] (SD)	211.5 (54.0)	216.6 (34.8)		163.3 (69.4)	151.5 (42.4)	171.4 (62.3)	

[a] Number of terminal hairs per cm^2 in the target area

SD standard deviation

decreased hair density, while 6 of 14 (43 %) sham-treated subjects did (Fig. 2g). In Trial #4, lasercomb-treated male subjects showed a higher percentage for hair density improvement of >5 hairs/cm^2 with either lasercomb model (86 % for the 9-beam model and 82 % for the 12-beam model) than the sham-treated subjects (59 %) (Fig. 2h). Whereas 9 of 22 (41 %) sham-treated subjects showed decreased hair density, only 3 of 21 (14 %) 9-beam lasercomb-treated subjects and 4 of 22 (18 %) 12-beam lasercomb-treated subjects did ($p = 0.0033$) (Fig. 2h).

Overall, we observed significant categorical improvement in terminal hair density with lasercomb treatment versus control (Fig. 2e–h). Taken together, all four trials using three different lasercomb models in both male and female subjects showed improvement in terminal hair density that was highly statistically significant, as well as categorical improvement, with lasercomb treatment compared with sham treatment at 26 weeks.

3.2.3 *Subject Self-Assessment*

A higher percentage of lasercomb-treated subjects reported overall improvement of hair loss condition and thickness and fullness of hair in self-assessment, compared with sham-treated subjects (Table 3). In Trial #1, statistical significance was reached for the assessment of both the overall improvement of hair loss condition and thickness and fullness of hair. Results in Trial #2 did not reach statistical significance. In the pooled male subject trials, assessment of the thickness and fullness of hair reached statistical significance, but not the overall improvement of hair loss condition (Table 3).

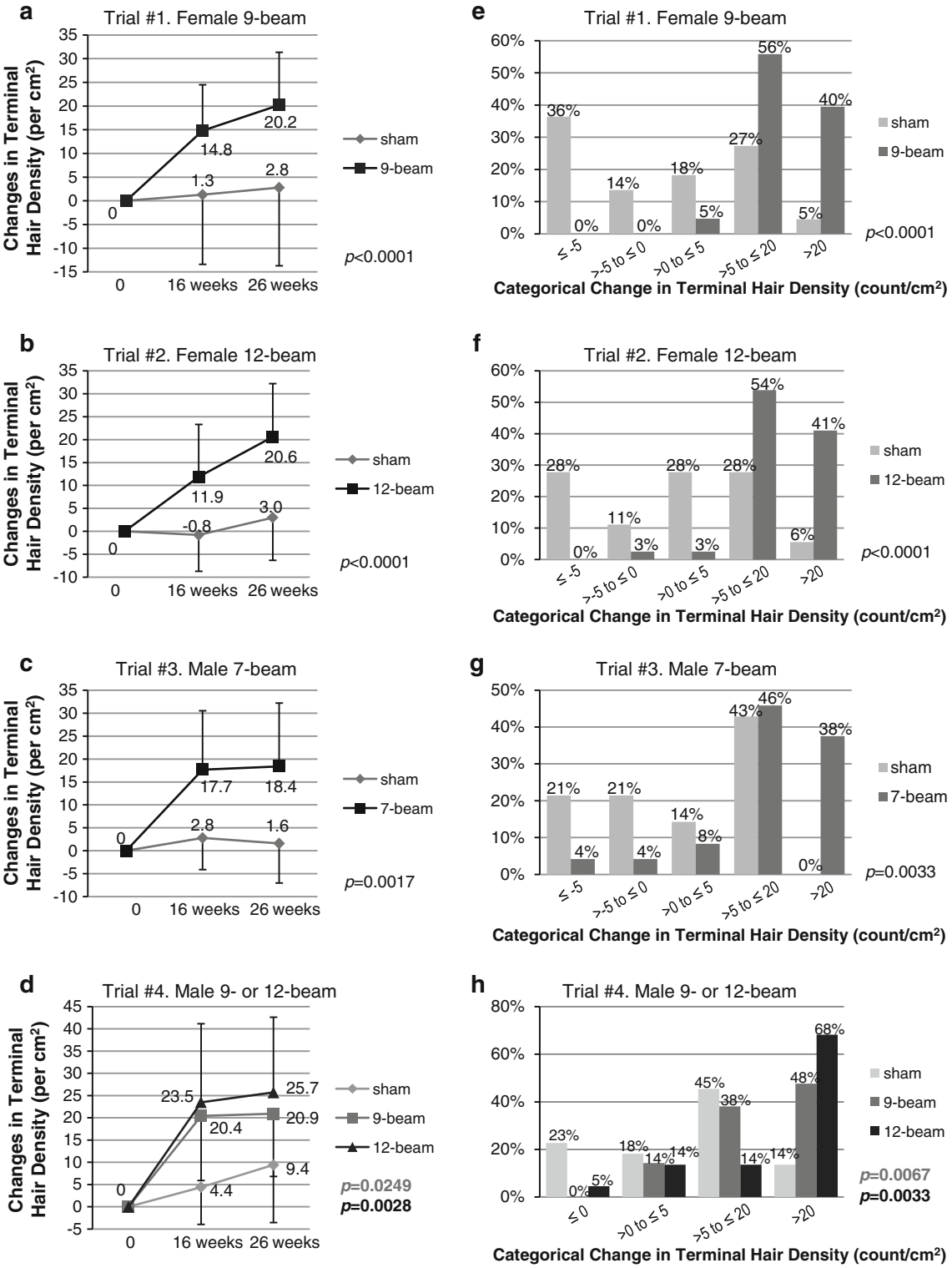

Fig. 2 **a**–**d** Mean changes in terminal hair density (count per cm^2) from baseline in subjects treated with the lasercomb or sham device. *Bars* indicate standard deviation. **e**–**h** Categorical changes in terminal hair density (count per cm^2) from baseline to 26 weeks in subjects treated with the lasercomb or sham device. Shown are *p* values at 26 weeks

Table 3 Subject self-assessment of efficacy (last observation carried forward)

	Trial #1. Female 9-beam				Trial #2. Female 12-beam				Trials #3 and #4. Male 7-, 9- and 12-beam (pooled)			
	Overall improvement of hair loss condition		Thickness and fullness of hair		Overall improvement of hair loss condition		Thickness and fullness of hair		Overall improvement of hair loss condition		Thickness and fullness of hair	
	Lasercomb ($n = 43$)	Sham ($n = 22$)	Lasercomb ($n = 43$)	Sham ($n = 22$)	Lasercomb ($n = 39$)	Sham ($n = 18$)	Lasercomb ($n = 39$)	Sham ($n = 18$)	Lasercomb ($n = 67$)	Sham ($n = 36$)	Lasercomb ($n = 67$)	Sham ($n = 36$)
Week 16 (LOCF), *n* (%)												
Improved/ minimally improved	36 (83.7)	12 (54.5)	28 (65.1)	8 (36.4)	25 (64.1)	7 (38.9)	23 (59.0)	8 (44.4)	39 (58.2)	12 (33.3)	37 (55.2)	15 (41.7)
No change	7 (16.3)	9 (40.9)	15 (34.9)	14 (63.6)	11 (28.2)	9 (50.0)	15 (38.5)	10 (55.6)	27 (40.3)	23 (63.9)	28 (41.8)	19 (52.8)
Worse/ minimally worse	0 (0)	1 (4.5)	0 (0)	0 (0)	3 (7.7)	2 (11.1)	1 (2.6)	0 (0)	1 (1.5)	1 (2.8)	2 (3.0)	2 (5.6)
	$p = 0.0149$*		$p = 0.0982$		$p = 0.0546$		$p = 0.2667$		$p = 0.0706$		$p = 0.2484$	
Week 26 (LOCF), *n* (%)												
Improved/ minimally improved	36 (83.7)	11 (50.0)	31 (72.1)	10 (45.5)	26 (66.7)	11 (61.1)	24 (61.5)	9 (50.0)	40 (59.7)	17 (47.2)	38 (56.7)	13 (36.1)
No change	6 (14.0)	11 (50.0)	12 (27.9)	12 (54.5)	11 (28.2)	6 (33.3)	14 (35.9)	7 (38.9)	25 (37.3)	16 (44.4)	28 (41.8)	21 (58.3)
Worse/ minimally worse	1 (2.3)	0 (0)	0 (0)	0 (0)	2 (5.1)	1 (5.6)	1 (2.6)	2 (11.1)	2 (3.0)	3 (8.3)	1 (1.5)	2 (5.6)
	$p = 0.0321$*		$p = 0.0345$*		$p = 0.1713$		$p = 0.1000$		$p = 0.0717$		$p = 0.0114$*	

* Statistically significant

LOCF last observation carried forward

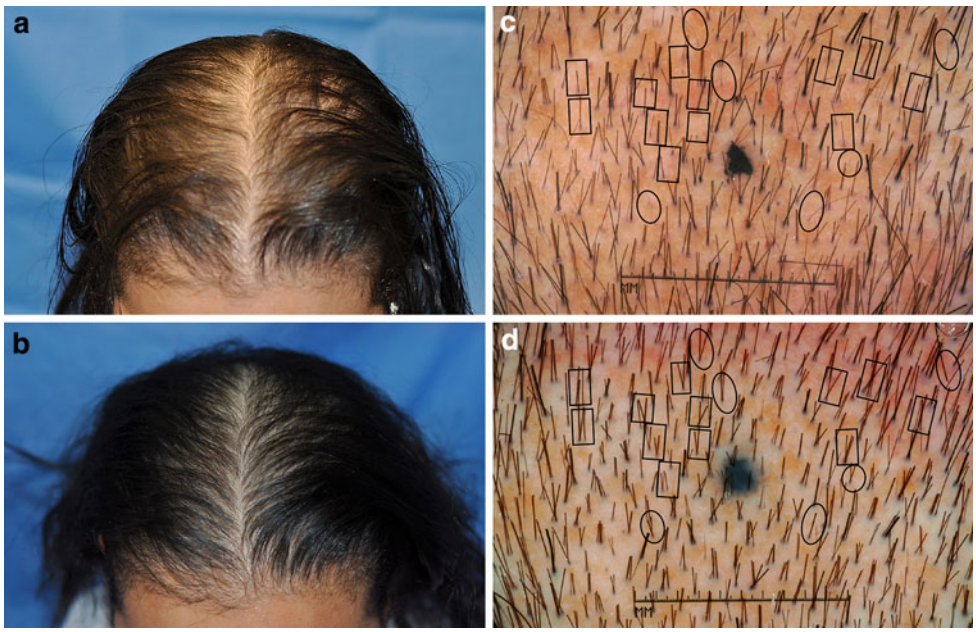

Fig. 3 Male and female pattern hair loss before and after lasercomb treatment. Global photographs of a female subject, at baseline (**a**) and after 26 weeks (**b**) of the 12-beam lasercomb treatment. Macrophotographs of a male subject, at baseline (**c**) and after 26 weeks (**d**) of the 9-beam lasercomb treatment. Increased hair count through conversion of vellus or intermediate follicles to active follicles producing terminal hair (*ovals*) or resting telogen to active anagen follicles (*rectangles*) is highlighted

3.2.4 Meta-Analysis of the Effects of Lasercomb Model, Study Duration, and Gender

Meta-analyses were conducted to provide an overall assessment of the individual study results. The overall results showed the least squares mean difference of change in terminal hair density of 15.27 (standard error 1.781) at 26 weeks from baseline between lasercomb- and sham treated subjects, which was highly statistically significant ($p < 0.0001$). The homogeneity assessment results were non-significant ($p = 0.6188$). These results indicated that compared with sham treatment, lasercomb treatment resulted in a statistically significant increase in terminal hair density across the trials, independent of the lasercomb model (7- and 9-beam 655 nm ± 5 % laser and 12-beam 635 nm and 655 nm ± 5 % laser) and the sex of the subject.

Before and after global photographs (Fig. 3a, b) and macrophotographs (Fig. 3c, d) demonstrated increases in terminal hair density, most likely through the conversion of vellus or intermediate follicles to terminal follicles or from resting telogen follicles to active anagen follicles.

In summary, efficacy analysis showed a statistically significant increase in terminal hair density after 26 weeks of lasercomb treatment compared with sham treatment. The mean increase in terminal hair density was higher (statistically significant) in lasercomb-treated subjects than in sham-treated subjects. Additionally, a higher percentage of lasercomb-treated subjects showed categorical hair density improvement (>5 hairs/cm^2) at 26 weeks, compared with sham-treated subjects. Such improvement was observed in all four trials, and independent of the sex and age of the subject, and independent of the lasercomb model when similar laser dose rates were delivered. A higher percentage of lasercomb-treated subjects reported overall improvement of hair loss condition and thickness and fullness of hair in self-assessment, though the results did not always reach statistical significance.

3.3 Safety and Tolerability

No serious adverse events were reported in any subject receiving the lasercomb in any of the four trials. Reported lasercomb-related adverse events included dry skin (5.1 %), pruritus (2.5 %), scalp tenderness (1.3 %), irritation (1.3 %), and a warm sensation at the site (1.3 %). No subjects experienced an adverse event that resulted in the discontinuation of the study device, or interruption of the study. No adverse events had an impact on the study device use. There were no significant differences in active device adverse events as recorded by device type.

4 Discussion

Pattern hair loss may affect up to 70 % of men and 50 % of women at some point in their lifetime [3, 4]. There has been an urgent need to determine whether LLLT home devices, which have been widely promoted for the treatment of MPHL and FPHL despite few randomized, controlled trials, can provide an effective alternative for patients with pattern hair loss, especially female patients. In this study, through four randomized, multicenter, sham device-controlled and double-blind clinical trials, we have shown that 26 weeks of treatment with the FDA-cleared HairMax LaserComb®, compared with sham treatment,

resulted in a statistically significant terminal hair density increase. Our results not only verified the effective treatment of MPHL reported previously [8], but also showed treatment efficacy in female subjects, and demonstrated that the treatment efficacy was independent of the laser configurations tested when similar laser dose rates were delivered. No serious adverse events were reported in any subject receiving lasercomb treatment in any of the four trials.

We have observed increased terminal hair density likely through both conversion of vellus or intermediate follicles to active follicles producing terminal hair and conversion of resting telogen follicles to active anagen follicles (Fig. 3c, d). The exact mechanisms of such conversions by LLLT remain unknown. Commonly used LLLT encompasses a wavelength of 500–1,100 nm and delivers fluences of 1–4 J/cm^2 with a power density of 3–90 mW/cm^2, and has demonstrated beneficial effects in various conditions including wound healing, joint pain relief, mucositis prevention and treatment, and skin conditions [15–22]. Based on anecdotal experience, LLLT of 650–900 nm wavelengths at 5 mW has been suggested to be an effective treatment option for male and female patients with pattern hair loss [23], though comprehensive studies evaluating laser modality are lacking. Whereas the exact mechanisms of hair growth stimulation by LLLT remain unknown, LLLT has been proposed to accelerate mitosis [24], and may stimulate HF stem cells or activate follicular keratinocytes. Additionally, laser light may alter cell metabolism through photodissociation of inhibitory nitric oxide from cytochrome c oxidase [25] (unit IV in the respiratory chain of mitochondria), causing increased ATP production and cellular activity [26]. Furthermore, resolution of inflammation may be one potential mechanism of hair growth stimulation by LLLT in AGA [27–32]. In vitro and in vivo trials of LLLT have shown decreased inflammatory prostaglandin E-2 [32] and proinflammatory cytokines [30], and in contrast, increased anti-inflammatory cytokines transforming growth factor-beta 1 and interleukin-10 [27, 28].

Results from the present investigation are consistent with the previous study of the 9-beam lasercomb in male AGA subjects by Leavitt et al. [8]. Both studies demonstrated a higher increase in terminal hair density with lasercomb treatment versus sham treatment, which was statistically significant, with a positive safety profile for the device. However, the current study enrolled both male and female subjects, and tested a range of laser configurations (8 min of treatment for the 12-beam model, 11 min for the 9-beam model, and 15 min for the 7-beam model, so that the three models gave similar laser dose rates per treatment), making it a more comprehensive study. While we found the lasercomb to be also efficacious in increasing terminal hair count in female subjects, we feel we cannot directly compare our results with another lasercomb study of female subjects ($n = 7$) as the baseline hair counts were too different (71–307/cm^2 vs. 8–32/cm^2) [9]. A recent study described the high efficacy of treating MPHL using a helmet-like low-level laser device, called TOPHAT®, in a randomized, double-blind, controlled trial [7]. While the TOPHAT® study was for 16 weeks with treatment every other day for a total of 60 treatments versus 78 treatments in total in this lasercomb study, the laser dose rates per treatment in the TOPHAT® study were much higher (there were 21 5-mW laser units). Future studies are required to optimize laser modality and treatment regimen for hair growth and maintenance.

The increase in terminal hair density per cm^2 observed in our study is comparable to that observed in a 6-month randomized, investigator-blinded, controlled trial of 5 % minoxidil solution in MPHL [33], but lower than that observed in 48-week studies of 5 % and 2 % minoxidil topical solution in MPHL [34] and FPHL [35]. Our results in the increase in terminal hair count are comparable to 1 mg/day finasteride treatment in some MPHL trials [36, 37], but less efficacious than longer term trials [38].

LLLT may provide a promising treatment option for patients who do not respond to either finasteride or minoxidil, and who do not want to undergo hair transplantation. Additionally, while topical minoxidil solution or foam is widely used to treat pattern hair loss and is generally well tolerated [39], the treatment needs to be applied once or twice daily, and be in contact with the scalp for at least 4 h. Such application can be impractical for many users, leading to noncompliance and reduced efficacy. As an alternative, the lasercomb treatment is safe and easy to apply, with 8–15 min of treatment three times per week, and leaves no residue on the scalp. Such user friendliness of the lasercomb may lead to better patient compliance and improved efficacy. Future studies to modulate laser modality and treatment regimen will help optimize hair growth stimulation and maintenance by low-level laser.

5 Conclusions

In four randomized, double-blind, sham-controlled trials of MPHL and FPHL, we detected a statistically significant increase in terminal hair density after 26 weeks of lasercomb treatment compared with sham treatment. Such improvement was independent of the sex and age of the subject, and independent of the lasercomb model when similar laser dose rates were delivered. A higher percentage of lasercomb-treated subjects reported overall

improvement of hair loss condition and thickness and fullness of hair in self-assessment, though the results did not always reach statistical significance. Increase in terminal hair count was comparable to the short-term trials of 5 % minoxidil topical solution and 1 mg/day finasteride, but less efficacious than longer term ($\geq$1 year) trials. Further clinical trials are needed to define the optimal duration of treatment, the duration of response, and the use of the lasercomb in other alopecia conditions.

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Quick Response Code:

Use of Low-Level Laser Therapy as Monotherapy or Concomitant Therapy for Male and Female Androgenetic Alopecia

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ABSTRACT

Background: Androgenetic alopecia (AGA) is the most common form of hair loss in men and in women. Currently, minoxidil and finasteride are the treatments with the highest levels of medical evidence, but patients who exhibit intolerance or poor response to these treatments are in need of additional treatment modalities. **Objective:** The aim was to evaluate the efficacy and safety of low-level laser therapy (LLLT) for AGA, either as monotherapy or as concomitant therapy with minoxidil or finasteride, in an office-based setting. **Materials and Methods:** Retrospective observational study of male and female patients with AGA, treated with the 655 nm-HairMax Laser Comb®, in an office-based setting. Efficacy was assessed with global photographic imaging. **Results:** Of 32 patients (21 female, 11 male), 8 showed significant, 20 moderate, and 4 no improvement. Improvement was seen both with monotherapy and with concomitant therapy. Improvement was observed as early as 3 months and was sustained up to a maximum observation time of 24 months. No adverse reactions were reported. **Conclusions:** LLLT represents a potentially effective treatment for both male and female AGA, either as monotherapy or concomitant therapy. Combination treatments with minoxidil, finasteride, and LLLT may act synergistic to enhance hair growth.

Key words: Androgenetic alopecia, concomitant therapy, HairMax Laser Comb®, low level laser therapy, monotherapy

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INTRODUCTION

The ability of lasers to induce hair growth was incidentally noted as early as 1967 when Mester *et al.* used low-level laser therapy (LLLT) to treat cancer in mice with shaved backs.[1] Since then, hypertrichosis has been recognized to be a possible side-effect of laser treatment. First described in 2002 with intense pulsed light therapy,[2] this phenomenon has now been widely acknowledged to occur with an incidence rate ranging from 0.6% to 10% with low fluences and all laser types.[3] It is thought to be the result of suboptimal fluences that are too low to induce thermolysis, but high enough to stimulate follicular growth.

Eventually, LLLT has been developed for the treatment of androgenetic alopecia (AGA). As opposed to other currently marketed systems, the laser comb utilizes hair parting teeth for optimal delivery of laser energy to the exposed scalp. In 2007, the HairMax Laser Comb® (Lexington International, LLC) received 510 (k) clearance from the Food and Drug Administration (FDA) for the treatment of AGA for men, and 2011 for women. This clearance means that the device is considered a moderate-risk medical device by the FDA and is thereby solely screened for safety. The HairMax Laser Comb® has been tested in a company-sponsored study of 110 male patients with the claim of a significant increase in mean terminal hair density when compared to a sham device.[4] Avram and Rogers conducted the first independent blinded study of LLLT and hair growth with seven patients and found that on average, there was a decrease in the number of vellus hairs, an increase in the number of terminal hairs, and an increase in shaft diameter.[5] A consensus written by hair loss experts states that based on anecdotal experience, LLLT, particularly 650-900 nm wavelengths at 5 mW, may be an effective treatment option for patients with AGA.[6] In recent times, Kim *et al.* reported an increase of hair density with the use of LLLT, when compared to the sham device in a 24-week, randomized, double-blind, sham-device-controlled trial.[7]

To evaluate efficacy of the 655 nm-HairMax Laser Comb® either as monotherapy or as concomitant therapy for

treatment of male and female AGA, we performed a retrospective observational study of global photographic assessments of patients in an office-based setting.

MATERIALS AND METHODS

The study design was retrospective and observational. Patients who had purchased a HairMax Laser Comb® between July 2011 and July 2013 for treatment of AGA at the Center for Dermatology and Hair Diseases Prof. Trüeb were retrieved for assessment of global photographic images performed at follow-up visits. Patients on concomitant treatment had been treating with topical minoxidil or oral finasteride for at least 9 months, before starting therapy with the HairMax Laser Comb®. Patients used the HairMax Laser Comb® at home according to instructions 3 times weekly between 8 and 15 min depending on the model purchased (Advanced 7, Lux 9, or Professional 12). Global photographs were performed at 3, 6, 12, and 24 months of treatment follow-up in a standardized manner with a stereotactic camera device of Canfield Scientific Inc., in which the patient's chin and forehead are fixed and on which digital camera and flash device are mounted, ensuring that view and lighting are the same at consecutive visits, thus enabling precise follow-up of the same scalp area of interest with frontal and vertex views. Global photographs were evaluated by two of the authors (AM and RMT), and scored as significant, moderate, or no improvement. In the case of diverging opinions, the inferior score was given.

RESULTS

In total, 32 patients with AGA were involved in the study, of which 21 were females, aged 22-73 (mean: 43.6 ± 15.19 standard deviation [SD]), and 11 were males, aged 20-70 (mean: 39 ± 15.01 SD) total mean: 42 ± 15.1 SD. The duration of hair loss in years for men and women was mean 7.1 ± 5.2 SD. The duration of LLLT in months for men and women was mean 8.7 ± 5.2 [Table 1]. The patient characteristics, with respect to gender, age, classification of AGA according to Ludwig and Hamilton-Norwood scales, duration of hair loss, and concomitant treatments are recorded in Table 2.

The results for the scoring of the global photographic assessment in relation to treatment duration with the HairMax Laser Comb® are demonstrated in Table 3. In summary, eight patients (three female, five male) showed significant improvement, 20 patients (14 female, six male) moderate improvement, and four patients (four female, zero male) no improvement [Figure 1]. Of 32 patients, the HairMax Laser Comb® was used as monotherapy in six patients (two female, four male), and as a concomitant therapy in 26 patients (19 female, seven male). In the monotherapy group, two patients (one female, one male) showed significant improvement [Figure 2], four patients (one female, three male) moderate improvement, and zero patients no improvement [Table 3]. In the concomitant therapy group, six patients (two female, four male) showed significant improvement [Figures 3 and 4], 16 patients (13 female, three male) moderate improvement, and four patients (four female, zero male) no improvement. There was no statistical significant difference between LLLT monotherapy and concomitant therapy with either minoxidil and/or finasteride ($P = 0.829$), and regarding male or female AGA ($P = 0.091$) [Table 4].

Treatment was well tolerated and no serious adverse events were reported.

DISCUSSION

Androgenetic alopecia is the most common form of hair loss in men and in women. Currently, topical 2% and 5% minoxidil solution and 1 mg oral finasteride are

Table 1: Improvement of alopecia in relation to the variables: Age, duration of hair loss, and duration of LLLT

Variables	Statistics	Total	Improvement			*P* value of Kruskal-Wallis test
			Number	Moderate	Significant	
Age (years)	*n*	32	4	20	8	0.381
	Mean	42.0	33.0	44.8	39.6	
	Standard deviation	15.1	6.8	16.4	13.5	
	Minimum	20.0	25.0	22.0	20.0	
	Maximum	73.0	40.0	73.0	62.0	
Duration of hair loss* (years)	*n*	24	4	13	7	
	Mean	7.1	7.3	7.0	7.4	0.892
	Standard deviation	5.2	3.9	5.8	5.6	
	Minimum	0.5	3.0	0.5	1.5	
	Maximum	20.0	11.0	20.0	16.0	
Duration of LLLT (months)	*n*	32	4	20	8	
	Mean	8.7	12.0	8.0	8.8	0.549
	Standard deviation	5.2	8.1	3.7	6.9	
	Minimum	2.0	6.0	2.0	3.0	
	Maximum	24.0	24.0	18.0	24.0	

LLLT – Low-level laser therapy

Table 2: Patient characteristics

Gender	Age	Classification	Duration of hair loss	Concomitant treatments
Male	25	Hamilton-Norwood III	NOS*	Nil**
Male	54	Hamilton-Norwood IV	20 years	Nil**
Male	34	Hamilton-Norwood IV	10 years	Nil**
Male	70	Hamilton-Norwood III	NOS*	Nil**
Male	28	Hamilton-Norwood IV	9 years	5% minoxidil solution
Male	32	Hamilton-Norwood IIIv	2 years	5% minoxidil solution
Male	56	Ludwig pattern	7 years	5% minoxidil solution
Male	20	Hamilton-Norwood IIIv	18 months	1 mg oral finasteride 1 mg+5% minoxidil solution
Male	34	Hamilton-Norwood IIIv	NOS*	1 mg oral finasteride 1 mg+5% minoxidil solution
Male	38	Hamilton-Norwood V	16 years	1 mg oral finasteride 1 mg+5% minoxidil solution
Male	38	Hamilton-Norwood IV	12 years	1 mg oral finasteride 1 mg+5% minoxidil solution
Female	73	Ludwig II	6 months	Nil**
Female	62	Ludwig I-II	2 years	Nil**
Female	71	Ludwig II	12 years	0.025% estradiol solution
Female	38	Ludwig II	NOS*	5% minoxidil solution
Female	31	Ludwig II	3 years	5% minoxidil solution
Female	39	Ludwig II	NOS*	5% minoxidil solution
Female	44	Ludwig I	15 years	5% minoxidil solution
Female	30	Ludwig I	10 years	5% minoxidil solution
Female	52	Ludwig II	3 years	5% minoxidil solution
Female	40	Ludwig I	3 years	5% minoxidil solution
Female	40	Ludwig I	30 months	5% minoxidil solution
Female	37	Ludwig I	3 years	5% minoxidil solution
Female	37	Ludwig I	5 years	5% minoxidil solution
Female	25	Ludwig I	11 years	5% minoxidil solution
Female	50	Ludwig I	4 years	5% minoxidil solution
Female	33	Ludwig II	8 years	5% minoxidil solution
Female	22	Ludwig I	NOS*	5% minoxidil solution
Female	24	Ludwig I	4 years	5% minoxidil solution
Female	69	Ludwig I	8 years	5% minoxidil solution
Female	45	Ludwig I	NOS*	5% minoxidil solution
Female	53	Ludwig I	NOS*	5% minoxidil solution

*NOS – Not otherwise specified; **NIL – Nothing

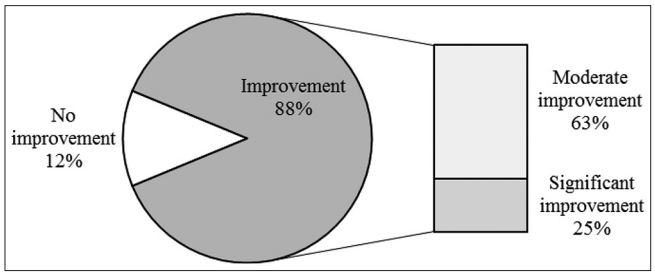

Figure 1: Graphic summary of results

the treatments with the highest levels of medical evidence,[8] but patients who exhibit intolerance or poor response to these treatments are in need of additional treatment modalities. Although low-level energy lasers have been therapeutically used in medicine for photobiostimulation in a variety of indications more than 30 years,[9] it has only recently found the attention of the scientific community for the treatment of AGA.[6,10,11]

We have chosen the 655 nm-HairMax Laser Comb® for several reasons: First, it represents the device with the most clinical study reports regarding its efficacy,[4,5,12] secondly, the cost of the device is affordable, and thirdly, the device is simple enough for patients to use at home. Finally, the fact that the device is safe, for which it received 510 (k) clearance from the FDA for the treatment of AGA, was also an important consideration.

Our study demonstrates clinical efficacy of the device for treatment of male and female AGA, both as monotherapy

Table 3: Scoring of global photographic assessment in relation to treatment duration

Gender	Age	Duration of LLLT	No improvement	Moderate improvement	Significant improvement
Male	25	4 months		X	
Male	54	12 months		X [Figure 2]	
Male	34	7 months			X
Male	70	7 months		X	
Male	28	4 months		X	
Male	32	6 months		X	
Male	56	3 months			X [Figure 3]
Male	20	10 months			X
Male	34	12 months		X	
Male	38	24 months			X [Figure 4]
Male	38	5 months			X
Female	73	2 months		X	
Female	62	06 months			X
Female	71	12 months		X	
Female	38	12 months			X
Female	31	3 months			X
Female	39	7 months		X	
Female	44	6 months		X	
Female	30	6 months	X		
Female	52	6 months		X	
Female	40	9 months	X		
Female	40	8 months		X	
Female	37	9 months		X	
Female	37	24 months	X		
Female	25	9 months	X		
Female	50	9 months		X	
Female	33	5 months		X	
Female	22	6 months		X	
Female	24	6 months		X	
Female	69	9 months		X	
Female	45	18 months		X	
Female	53	12 months		X	

LLLT – Low-level laser therapy

Table 4: Comparative assessment of efficacy between monotherapy and concomitant for male and female androgenetic alopecia

	Total (*n* (%))	Improvement (*n* (%))			*P* value of Fisher test
		Number	Moderate	Significant	
Gender					
Male	11 (34.4)	0	6 (30.0)	5 (62.5)	0.091
Female	21 (65.6)	4 (100.0)	14 (70.0)	3 (37.5)	
Therapy					
Monotherapy	6 (18.8)	0	4 (20.0)	2 (25.0)	0.829
Concomitant therapy	26 (81.3)	4 (100.0)	16 (80.0)	6 (75.0)	

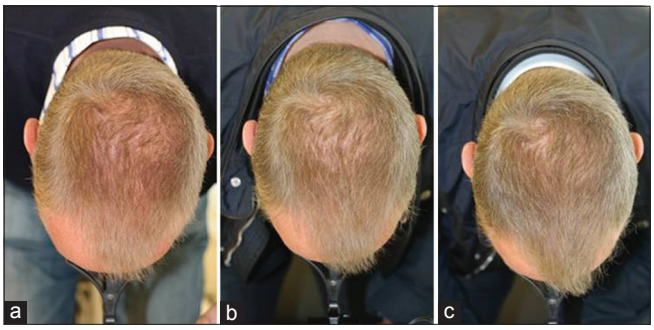

Figure 2: Monotherapy in a 54-year-old male (a) Before treatment, and improvement after (b) 6 months, and (c) 12 months of low-level laser therapy

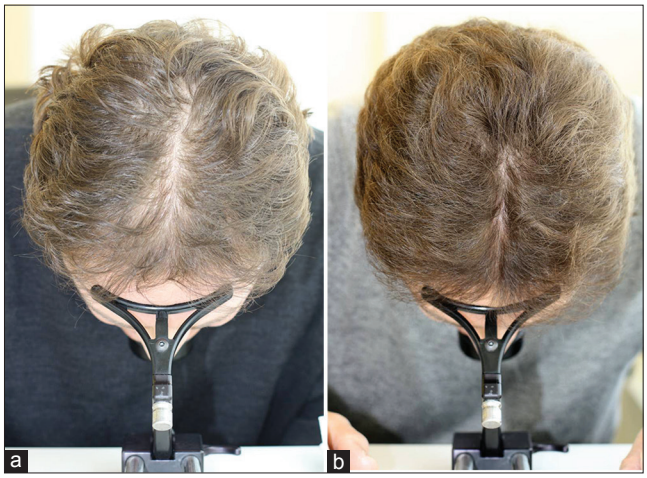

Figure 3: Concomitant treatment with topical 5% minoxidil in a 55-year-old male adding on low-level laser therapy (LLLT) to 4 year pretreatment with 5% topical minoxidil solution (a) Before, and (b) After 3 months of added LLLT

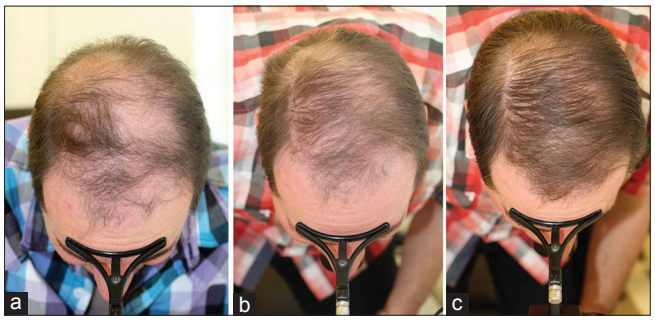

Figure 4: Concomitant treatment with topical 5% minoxidil and 1 mg oral finasteride in a 34-year-old male (a) Before, (b) After 9 months treatment with 1 mg oral finasteride and topical 5% minoxidil solution bid, and (c) After 3 months after adding on low-level laser therapy

and as concomitant therapy, in terms of clinically relevant improvement of appearance of hair. Of 32 patients, eight patients (25%) showed significant improvement, and 20 patients (62.5%) showed moderate improvement in global photographic assessments. The effect was observed

as early as 3 months of treatment, and was sustained up to a maximum observation time of 24 months. The technology appears to work better for some than for others, and predictive factors which will most benefit from LLLT are to be determined. It seems though, that patients with intermediate alopecia (Hamilton-Norwood III and IV, and Ludwig I and II, respiratory) respond best, since effective photobiostimulation depends on a minimum of hair for effective photobiostimulation, and on a maximum of hair for the laser beam to reach the scalp without absorption or interference from existing hairs.

The hypothesized mechanisms of action of LLLT are increased adenosine tri-phosphate (ATP) production, modulation of reactive oxygen species (ROS), and induction of transcription factors. The proposed cellular chromosphere responsible for the effect of visible light is cytochrome c oxidase (COX) with absorption peaks in the near infrared, and mitochondria the likely site for the initial effects. It is believed that LLLT displaces nitric oxid from COX allowing an influx of oxygen to bond to COX and progress forward in the respiratory process to ATP production and ROS signaling. These effects in turn lead to increased cellular proliferation, modulation in levels of cytokines, growth factors and inflammatory mediators, and increased tissue oxygenation. While the effects of these biochemical and cellular changes have broadly been studied in both animal models and clinical studies with patients, and have shown benefits in diverse conditions, such as increased healing in chronic wounds, improvements in sports injuries and carpal tunnel syndrome, pain reduction in arthritis and neuropathies, and amelioration of damage after heart attacks, stroke, nerve injury and retinal toxicity,[7,9] the effects on hair growth stimulation have only recently gained the attention of the scientific community.

CONCLUSIONS

From our own observations, we share with other authors the opinion that LLLT represents a safe and potentially effective treatment option for patients with AGA who do not respond or are not tolerant to standard treatment of AGA.[6,7] Moreover, combining LLLT with topical minoxidil solution and oral finasteride may act synergistic to enhance hair growth. Due to the known beneficial effect on wound healing, it is conceivable that LLLT as an adjunctive therapy in hair transplant surgery may also reduce postoperative shedding, reduce healing time, and increase graft patency. The scientific basis for such an approach is given, but there is a need for controlled studies with a higher number of patients to establish an increase in efficacy of combination regimens.[13]

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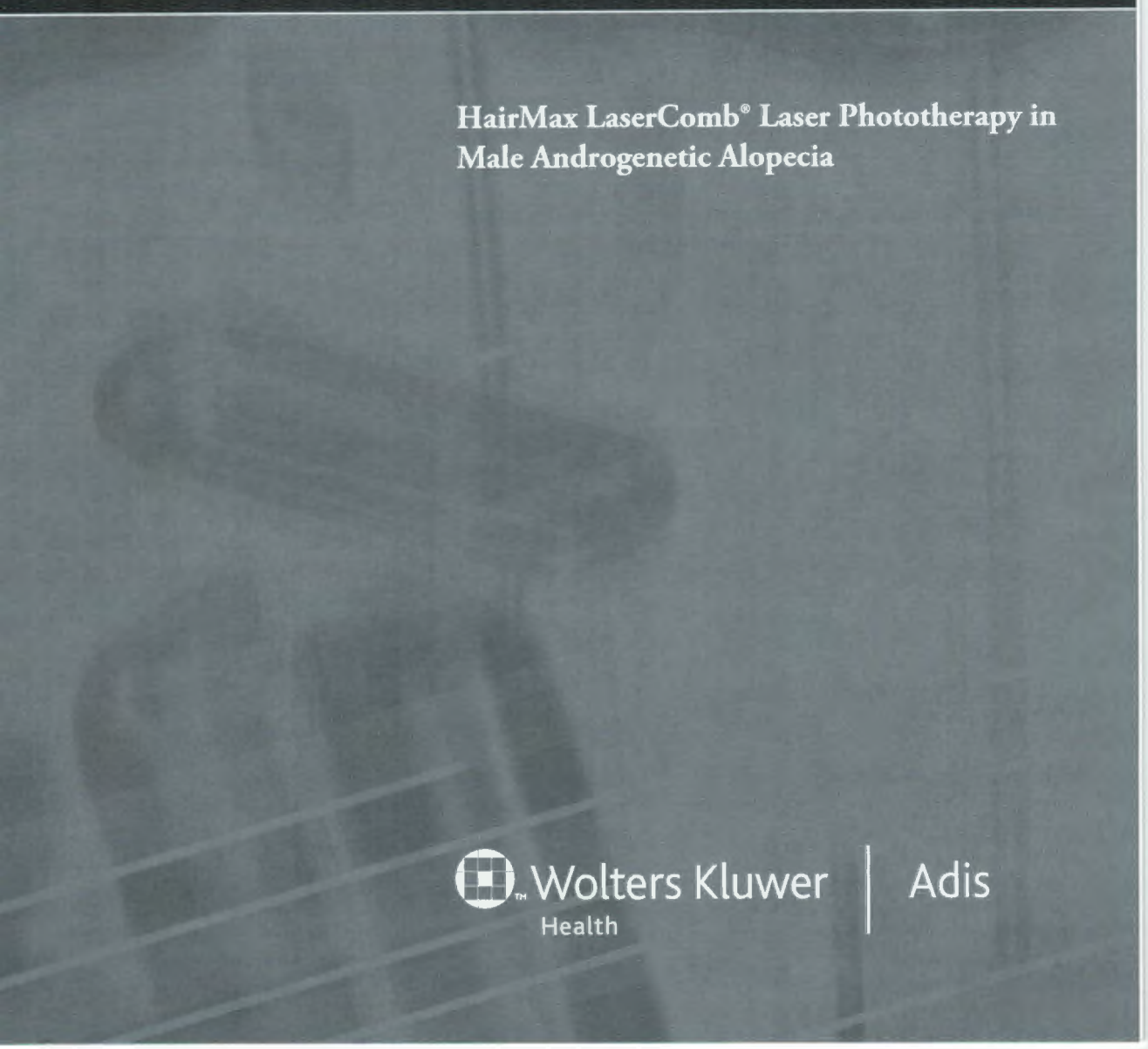
HairMax LaserComb® Laser Phototherapy in Male Androgenetic Alopecia



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HairMax LaserComb® Laser Phototherapy Device in the Treatment of Male Androgenetic Alopecia

A Randomized, Double-Blind, Sham Device-Controlled, Multicentre Trial

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Abstract

Background and objective: The use of low levels of visible or near infrared light for reducing pain, inflammation and oedema, promoting healing of wounds, deeper tissue and nerves, and preventing tissue damage has been known for almost 40 years since the invention of lasers. The HairMax LaserComb® is a hand-held Class 3R lower level laser therapy device that contains a single laser module that emulates 9 beams at a wavelength of 655 nm (±5%). The device uses a technique of parting the user's hair by combs that are attached to the device. This improves delivery of distributed laser light to the scalp. The combs are designed so that each of the teeth on the combs aligns with a laser beam. By aligning the teeth with the laser beams, the hair can be parted and the laser energy delivered to the scalp of the user without obstruction by the individual hairs on the scalp. The primary aim of the study was to assess the safety and effectiveness of the HairMax LaserComb® laser phototherapy device in the promotion of hair growth and in the cessation of hair loss in males diagnosed with androgenetic alopecia (AGA).

Methods: This double-blind, sham device-controlled, multicentre, 26-week trial randomized male patients with Norwood-Hamilton classes IIa–V AGA to treatment with the HairMax LaserComb® or the sham device (2 : 1). The sham device used in the study was identical to the active device except that the laser light was replaced by a non-active incandescent light source.

Results: Of the 110 patients who completed the study, subjects in the HairMax LaserComb® treatment group exhibited a significantly greater increase in mean terminal hair density than subjects in the sham device group ($p < 0.0001$). Consistent with this evidence for primary effectiveness, significant improvements in overall hair regrowth were demonstrated in terms of patients' subjective assessment ($p < 0.015$) at 26 weeks over baseline. The HairMax LaserComb® was well tolerated with no serious adverse events reported and no statistical difference in adverse effects between the study groups.

Conclusions: The results of this study suggest that the HairMax LaserComb® is an effective, well tolerated and safe laser phototherapy device for the treatment of AGA in males.

Background

Laser phototherapy is a popular therapeutic modality that relies on exposure of biological tissues to low power coherent monochromatic light;[1] this induces a variety of positive therapeutic benefits associated with a range of wavelengths from red through to infrared. Pioneer studies on laser biostimulation performed more than 40 years ago reported a prominent hair growth stimulatory effect in mice.[2] In recent years, considerable attention has been given to establishing a regeneration-promoting effect of laser phototherapy in wound healing and tendon, muscle,[3] fractured bone[4,5] and skin.[6] Most prominently, several recent studies have confirmed the stimulatory effect of laser phototherapy on cutaneous wound regeneration (i.e. wound healing).[6-10] Stimulation of proliferation was found to be at least one of the mechanisms underlying the pro-regenerative effect of laser phototherapy.[11-13] Because both reparative regeneration, which occurs during wound healing, and physiological regeneration, which occurs during the hair cycle, rely heavily on cell proliferation, it is plausible to suggest that the hair growth stimulatory effect of laser phototherapy is also mediated through either a direct or an indirect increase in proliferative activity within the hair follicle epithelial matrix.

The basis of the biostimulatory effect of laser phototherapy during wound healing is not fully understood. As noted above, at the cellular level, laser phototherapy has been shown to increase proliferation of fibroblasts,[11-13] including fibroblasts derived from streptozotocin-diabetic rats that otherwise exhibit impaired proliferative activity.[14] Several intracellular processes are believed to underlie this pro-proliferative effect, including short-term activation of the mitochondrial electron-transport chain, accumulation of intracellular adenosine triphosphate and alkalization of the cytoplasm.[11,15] Because laser phototherapy-promoted wound healing is also characterized by faster wound re-epithelialization and neovascularization,[14] direct enhancement of epidermal and endothelial proliferation in wound sites is plausible. In addition, the pro-proliferative action of laser phototherapy can be attributed to other indirect effects, one of which is a 'metabolic boost' of the regenerating tissues through increased cutaneous microcirculation that occurs upon laser irradiation.[16] Another effect is linked to stimulated secretion of endogenous growth factors, such as basic fibroblast growth factor and insulin-like growth factor-1, by fibroblasts exposed to laser phototherapy.[17] Both of these growth factors are potent natural stimulators of proliferation for a variety of cell types.[17]

Hair is one of the fastest growing tissues of the human body. Hair follicles undergo repetitive physiological regenerative cycles,[18] and each such cycle consists of three principal phases: telogen (resting phase), anagen (active phase) and catagen (physiological involution phase). At the basis of this hair growth cycle are two major processes. The first represents tightly controlled activation of epithelial bulge stem cells and secondary hair germ cells that give rise to transient amplifying (TA) progeny cells during telogen-to-anagen transition.[19] The second process constitutes robust proliferation of these TA cells within the epithelial matrix of the hair follicle throughout the entire length of anagen. Proliferation trichocytes terminally differentiate to form the bulk of the hair filament – the final product of the hair cycle. The dermal papilla of the hair follicle is believed to play a key regulatory role in orchestrating the above described processes of progenitor cell activation, hair matrix cell proliferation and terminal differentiation of trichocytes.[20]

Androgenetic alopecia (AGA) is one of the most common forms of hair loss in males and females.[21] In genetically predisposed scalp hair follicles, dihydrotestosterone – a potent derivative of the male

sex hormone testosterone – initiates the cascade of downstream signalling changes beginning in the dermal papillae fibroblasts that ultimately disturb normal metabolic and cellular dynamics of the entire follicle.[22] As a result, a marked reduction in proliferative activity in the hair follicle epithelium leads to morphological miniaturization of terminal scalp hairs into vellus-like hairs.[23] Furthermore, a broken mechanism of bulge stem cell and secondary hair germ cell activation prevents new anagen re-entry, converting cycling hair follicles into quiescent telogen follicles. Thus, while the aetiological basis of AGA is clearly in abnormal androgen signalling, disruption of epithelial progenitor cell activation and TA cell proliferation forms an essential pathophysiological component of this condition.[23]

Since laser phototherapy has pro-proliferative effects in a variety of tissues and cell types, we hypothesized that it might have similar pro-proliferative activity in hair follicles and might normalize physiological regeneration of scalp hair follicles affected in AGA. The phenomenon of so-called 'terminalization' of vellus human hair follicles (i.e. when small vellus hairs transform into larger, terminal hairs) upon low fluence diode laser treatment has been independently reported by two researchers.[24,25]

To further evaluate the validity of our assumptions, we measured the hair growth-promoting efficacy of the HairMax LaserComb® laser phototherapy device in a randomized, double-blind, sham device-controlled, multicentre trial in male patients with AGA. The HairMax LaserComb® is a handheld Class 3R lower level laser therapy device that contains a single laser module that emulates 9 beams at a wavelength of 655 nm (±5%). From past 'in-use' experience with the first devices it was found that there is a so-called 'optical window' for lower level light (LLL) in skin. LLL in skin appears to be effective in red and near-infared spectrum (600–950 nm) and the HairMax LaserComb® was found to be optimally effective at a wavelength of 655 nm (±5%).[26] The device uses a technique of parting the user's hair by combs that are attached to the device. This improves delivery of distributed laser light to the scalp. The combs are designed so that each of the teeth on the combs aligns with a laser beam. By aligning the teeth with the laser beams, the hair can be parted and the laser energy delivered to the scalp of the user without obstruction by the individual hairs on the scalp. Here we report on the outcome of this trial.

Methods

This clinical study was performed in accordance with Good Clinical Practice. The protocol was approved by the Investigational Review Board, Research Testing Laboratories Inc., Great Neck, NY, USA, and written informed consent was obtained from each patient in the study before study procedures were conducted.

Study Objectives

The primary aim of the study was to assess the safety and effectiveness of the HairMax LaserComb® laser phototherapy device in the promotion of hair growth and in the cessation of hair loss in males diagnosed with AGA. After the study began it was amended to include only males at the suggestion of the US FDA; female subjects will form the basis of a similar study.

Study Inclusion/Exclusion Criteria

For inclusion in the study, subjects must not have used or taken any of the following medications for 6 months prior to initiation of the study: minoxidil, finasteride (or any other 5α-reductase inhibitor medications), medications with anti-androgenic properties (e.g. cyproterone, spironolactone, ketoconazole, flutamide and bicalutamide), topical estrogens, progesterone, tamoxifen, anabolic steroids, medications that can potentially cause hypertrichosis (e.g. ciclosporin, diazoxide, phenytoin and psoralens), oral glucocorticoids (inhaled glucocorticoids were permitted), lithium, phenothiazines or other medications at the discretion of the investigator. Subjects were excluded if they had had hair transplantation, scalp reduction, current hair weave or tattooing of the alopecic area, which would have made for difficulties in performing objective hair

density assessments. Subjects were also excluded if they had any known underlying medical conditions that could adversely affect hair growth, such as HIV infection, connective tissue disease, inflammatory bowel disease, or other pathologies at the discretion of the investigator.

Patient Population

The study population included males between the ages of 30 and 60 years with a diagnosis of AGA who had been experiencing progressive hair loss within the last 12 months. Subjects were also required to have a Norwood-Hamilton male pattern hair loss classification of IIa to V and to have skin type I to IV on the Fitzpatrick Skin Type Scale.

Study Design

The study was designed as a randomized, double-blind, sham device-controlled, multicentre trial conducted at four sites in the US. Subjects who met all entry criteria received either the HairMax LaserComb®, which emitted laser light, or a sham device, which was identical to the active device in appearance but emitted incandescent light instead.

Screened subjects who fulfilled study entry criteria attended a baseline visit. At this visit, medical personnel assessed the subject's scalp for any signs of irritation or dermatological conditions that would disqualify the subject from participation. All subjects had systolic and diastolic blood pressure (mmHg) and heart rate (beats/min) vital signs recorded. Scalp macroimages utilizing dot mapping and computer-aided hair counts (see Evaluation of Clinical Efficacy section) were taken to document hair loss progression since the screening visit. Each target site for investigation was chosen by the clinical investigator based on the appearance of miniaturized hairs, which are the hallmark of AGA. Target scalp areas were identified and tattooed, then clipped to determine baseline hair density. Subjects were then provided with their randomized HairMax LaserComb® or sham device (see Statistical Analysis section for randomization scheme). In previous 'in-use' studies utilizing various application regimens to find the effective minimum dose of the HairMax LaserComb®, it was found that application of the device three times a week was sufficient to induce hair growth.[26] Therefore, subjects were asked in the current study to use the device assigned three times per week for 15 minutes on non-concurrent days for a total of 26 weeks/6 months. Subjects were given diaries to document use of the device.

Subjects returned to the clinic at 8 and 16 weeks to undergo assessment for adverse events and concomitant medications, collection of vital signs, scalp evaluation for local dermatitis and other pathological conditions, and completion of an 11-item questionnaire. Clinical assessment of treated scalp sites was carried out objectively at 26 weeks/6 months utilizing macroimaging techniques, hair clippings, computer-aided hair counts (see Evaluation of Clinical Efficacy section) and global assessment of new hair growth (i.e. without referring to any macroimages) by subjects and the investigator. Subjects who terminated prematurely had their hair density measured at their termination visit.

The evaluator of the baseline and endpoint analyses of the macroimages used blinded patient files and was not involved in patient selection or distribution of either device. The cut-off time for use of the device was 6 months. At the completion of the study, all subjects in each arm of the study were offered a HairMax LaserComb® for their personal use.

No post-study follow-up was conducted.

Evaluation of Clinical Efficacy

Hair Clipping

A circle of approximately 2.96 cm diameter, positioned in the vertex portion of the scalp, was identified as the site for hair clipping. This site contained some miniaturized hairs and was the target area for the hair density evaluation. A template was provided to the investigator for identifying the area for hair clipping. Once the hair had been clipped, trained study personnel used a professional tattooing machine to apply a permanent ink dot, approximately the size of a full stop/period (.), in the centre of the circle. The tattoo was used as a guide for placing the template on the scalp surface at

subsequent visits for the hair clipping and macroimages for hair density evaluation.

Macroimaging and Hair Density Evaluation Procedures

Macroimage Acquisition

The subject sat in a chair and was correctly placed in the stereotactic apparatus. The digital images were standardized for lighting, camera angle and position of the subject's head in each digital image to achieve a similar camera angle and relative image size. For macrodigital images a template as described in the previous section was placed on the lenses for precise and consistent alignment on the tattoo. A 10 mm scale divided into 0.1 mm increments was etched into the template for calibration purposes during the hair density evaluations. The images were recorded on compact flash cards. During the subject's visit, the images were previewed to see if they were acceptable; unacceptable images were retaken. After the subject's visit had been completed, the images were printed and signed by the investigator and uploaded to a designated site for image archiving.

Image Analysis and Management

A state-of-the art software system was utilized for image management. Macroimages were imported into a blinded subject file labelled by subject number. The images displayed included a means of marking each individual hair. Each hair was 'clicked' and a running count was displayed at the bottom of the software window. Only terminal hairs were counted. Archives of the counted hairs were maintained in the subject file. Images could be displayed side by side.

The database software functionality also allowed subjects to be identified by number while the advanced multiple criteria searches facilitated quick retrieval of information. In addition, the subject's chart view allowed all of a subject's images to be viewed on one screen with scalable thumbnails. Blinded subject records containing hair density measurements were forwarded to data management for inclusion in the study data base.

Study Endpoints

The primary efficacy endpoint was change in non-vellus terminal hair density (hairs/cm^2) in the target region between baseline and endpoint (26 weeks/6 months or the earlier termination visit), as assessed by scalp macroimaging using dot mapping and computer-aided hair counts (see Evaluation of Clinical Efficacy section). The techniques used were comparable to those used in the protocols for minoxidil.

Secondary effectiveness endpoints were subjective global assessment of hair regrowth by subjects and the investigator at week 26. Patients were asked to complete the 11-question proprietary Subject Questionnaire without assistance, aimed at evaluating perception of overall hair regrowth characteristics. These questions encompassed overall results, rate of hair loss, assessment of dandruff, scalp health, hair health, hair thickness, hair shine, hair growth rate, manageability and hair colour changes. Investigators were asked to evaluate the subject's hair growth looking at the baseline and week 26/6 months (or early termination) visit global images (without reference to macroimages) and graded the hair growth on a 4-point scale.

The safety endpoints for the study were adverse events of any nature and vital signs.

Statistical Analysis

Based on prior data for the HairMax LaserComb®[26] and the drug minoxidil (NDA 20-834, Pharmacia and Upjohn Consumer Healthcare, November 14, 1999) the standard deviation of change from baseline in terminal hair density was assumed to be 30 hairs/cm^2. Based on this estimate, 93 subjects randomized 2:1 (62 in the HairMax LaserComb® group and 31 in the sham device group) would provide 85% power to detect a difference of 20 hairs/cm^2. To allow for a 20% dropout rate, 123 subjects needed to be enrolled. Statistical comparisons were made between treatment groups for all baseline demographic variables. Continuous variables were compared using two-sample t-tests: dichotomous variables were compared using Pearson's chi-squared test (χ^2) and ordinal variables by

density assessments. Subjects were also excluded if they had any known underlying medical conditions that could adversely affect hair growth, such as HIV infection, connective tissue disease, inflammatory bowel disease, or other pathologies at the discretion of the investigator.

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the Cochran-Mantel-Haenszel procedure after assignation of uniform scores (1, 2, 3, etc.) to the four ordered categories of response.

The primary analysis of effectiveness was performed on all randomized subjects who had a post-baseline hair density measurement. The last value was carried forward for subjects who terminated prematurely. All randomized subjects who used the study device at least once were included in analyses of safety. The primary analysis of effectiveness was an analysis of covariance (ANCOVA), which included the effects of treatment group, study centre, age (as a continuous variable), and Fitzpatrick Skin Type Scale classification (as a categorical variable with four levels).

Adverse events were summarized as the number and percentage of subjects reporting each event. Statistical comparisons were made between treatment groups using Fisher's exact test.

All statistical analyses were two-sided at a 5% level of significance.

Results

Study Population

A total of 123 subjects were enrolled at four study sites. Table I shows the mean age, race and Fitzpatrick Skin Type Scale classification of subjects entered into the study. Seven subjects were discontinued from the study by the sponsor because of deviations from baseline entry criteria. This was because the site location chosen for the target area for hair density evaluation was found to be outside the zone of miniaturized hairs. The study design required that chosen sights for evaluation had to have miniaturized hairs. One subject was discontinued because of noncompliance with study visits. One subject was lost to follow-up. Four subjects withdrew consent for other reasons. Of these four subjects, two subjects in the sham device group had early termination visits (at 71 and 112 days) at which hair density measurements were completed. Ten subjects in the HairMax LaserComb® group who terminated prematurely were not included in the primary analysis of effectiveness, and one of the three subjects in the sham device group who terminated prematurely was not included.

Table I. Baseline demographics of the study population (males, n = 123)

Characteristic	Value
Age (y)	
Mean ± SD	47.9 ± 8.7
Range	30–60
Race [n (%)]	
White, non-Hispanic	111 (90.2)
Hispanic	9 (7.3)
Black	0 (0)
Other	3 (2.4)
Fitzpatrick Skin Type Scale classification [n (%)]	
I	4 (3.3)
II	17 (13.8)
III	65 (52.9)
IV	37 (30.1)

Primary Efficacy

As noted previously, hair counts were performed utilizing macroimages imported into blinded patient files by an evaluator who was not connected with the clinical trial. The two subjects with the greatest decreases in hair density (subject 04-039 in the sham device group with −145 hairs/cm^2 and subject 01-039 with −56 hairs/cm^2) appeared to be outliers in the statistical analysis. The residual standard deviation was used as an estimate of the accuracy of the dependent variable being measured, hair density. The ANCOVA was 18.6 hairs/cm^2 and these subjects had residuals of −128.0 (subject 04-039) and −75.8 (subject 01-039). To assess the impact of these subjects on analysis, they were removed and the results are shown in table II. Removal of these subjects reduced the residual standard deviation from 18.6 to 11.2 hairs/cm^2; however, the impact of the removal of these two outlier subjects on the final results was negligible.

When the two outliers were excluded from the analysis, subjects treated with the HairMax LaserComb® had a mean increase in terminal hair density of +19.8 hairs/cm^2, while subjects in the sham device group had a mean decrease of −7.6

Table II. Mean baseline and change from baseline to 26 weeks in terminal hair density[a] (hairs/cm²) – two outliers excluded

Time	HairMax LaserComb® (n = 71)	Sham device (n = 39)
Baseline		
mean ± SD	122.9 ± 51.4	120.7 ± 48.6
range	21.6, 252.1	25.5, 225.4
Change from baseline		
mean ± SD	17.3 ± 11.9	−8.9 ± 11.7
range	−6.4, 52.2	−54.7, 7.6
Adjusted mean[b]	19.8	−7.6
Difference (95% CI)	27.4 (22.9, 31.9)	
p-Value	<0.0001	

a Last value carried forward for subjects who terminated prematurely.

b Adjusted for study centre, subject's age and skin type.

hairs/cm² at the completion of the study (table II). This difference was significant ($p < 0.0001$). An example of terminal hair regrowth in the non-vellus hair density macroimages of one patient in the HairMax LaserComb® group is shown in figure 1.

Table III shows individual subject changes from baseline in terminal hair density, divided into six categories. Only two subjects in the HairMax LaserComb® group (2.8%) had a decrease in hair density ≥5 hairs/cm², whereas 26 subjects in the sham device group (65.0%) had a similar decrease. Furthermore, 62 subjects in the HairMax LaserComb® group (86.1%) had an increase in hair density >5 hairs/cm², while only two subjects in the sham device (5.0%) group had such an increase. Significant improvements in overall hair regrowth were demonstrated in terms of patients' subjective assessment ($p < 0.01$) at 26 weeks over baseline.

Secondary Efficacy Analyses

Secondary effectiveness endpoints included subjects' assessment of overall hair growth (table IV), the investigator's global assessment of overall hair growth (table IV), and responses to ten additional questions in the Subject Questionnaire. In each of the following analyses, subjects who terminated prematurely had their last value carried forward to each subsequent visit.

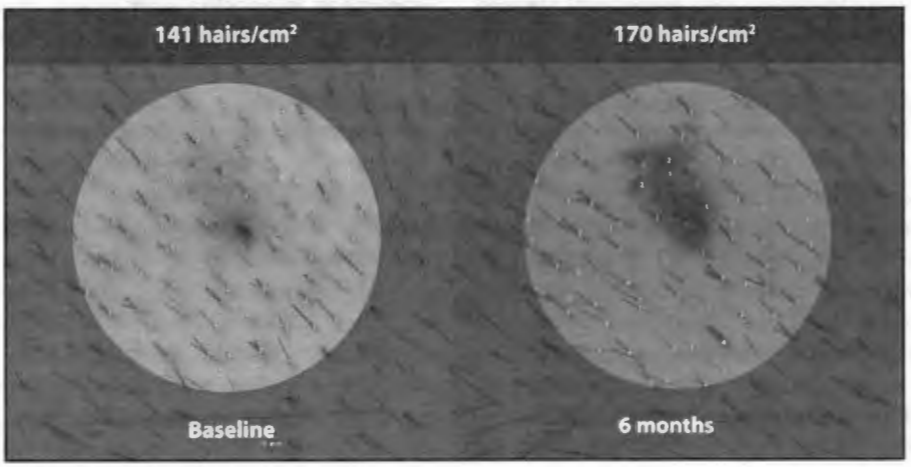

Fig. 1. Non-vellus hair density macroimages at baseline and 6 months in one patient in the HairMax LaserComb® group (6-month image shows evidence of ink spread).

Table III. Categorical changes from baseline to 26 weeks in terminal hair density[a]

Change in hair density/cm²	HairMax LaserComb® (n=72) [n (%)]	Sham device (n=40) [n (%)]
≤–20	1 (1.4)	7 (17.5)
≥–20 to –5	1 (1.4)	19 (47.5)
≥–5 to 0	3 (4.2)	9 (22.5)
>0 to 5	5 (6.9)	3 (7.5)
>5 to 20	34 (47.2)	2 (5.0)
>20	28 (38.9)	0 (0)

a Last value carried forward for subjects who terminated prematurely.

As seen in table IV showing subjects' assessment of overall hair growth, the p-value for the comparison between treatment groups achieved statistical significance (p=0.01) at the last visit. Thus, subjects in the HairMax LaserComb® group perceived significantly greater improvement in hair regrowth than those in the sham device group at the end of the study.

The results of the investigator's global assessments of hair growth are also shown in table IV. No substantial differences were seen between treatment groups within each assessment category (p=0.45). The discordant results between the investigator's subjective global assessment shown in this table and the objective hair density measurements shown in table II were assessed by comparing the median changes from baseline in terminal hair density within each treatment group and category of response. Medians are used rather than means to lessen the impact of large decreases. This assessment showed that there was no correlation between median changes from baseline in actual terminal hair density shown in table II and investigator subjective global assessment shown in table IV among subjects in the HairMax LaserComb® group. These results question the validity of the investigator's subjective global assessment because of their lack of any agreement with actual terminal hair growth shown in table II. It also brings into question the validity of global photography of hair due to the numerous inherent variables that effect the appearance of hair at each evaluation point.

Of the ten additional questions remaining in the end-of-study Subject Questionnaire after subjects' assessment of overall hair regrowth (table IV), answers to seven were analysed statistically (three questions – reduced dandruff, return to natural colour, and scalp irritation – were excluded because of high proportions of 'not applicable' answers). For the remaining seven questions, the responses to five (slower hair loss, better scalp health, thicker feeling hair, more shine to hair and overall hair improve-

Table IV. Subjects' and investigator's assessment of overall hair regrowth at week 26

Assessment	HairMax LaserComb® [n (%)]	Sham device [n (%)]	p-Value[a]
Subjects' assessment at week 26	**n=76**	**n=39**	
No growth	28 (36.8)	21 (53.9)	0.01
Minimal growth	30 (39.5)	16 (41.0)	
Moderate growth	17 (22.4)	2 (5.1)	
Dense growth	1 (1.3)	0 (0)	
Investigator's assessment at week 26	**n=72**	**n=38**	
No growth	46 (63.9)	22 (57.9)	0.45
Minimal growth	18 (25.0)	10 (26.3)	
Moderate growth	7 (9.7)	5 (13.2)	
Dense growth	1 (1.4)	1 (2.6)	

a HairMax LaserComb® vs sham device.

ment) were statistically significantly different ($p < 0.05$) between the groups, i.e. the assessments were significantly better in subjects in the HairMax LaserComb® group compared with those in the sham device group. Although between-group differences in responses to the last two questions (faster growing hair, more manageable hair) did not achieve statistical significance ($p < 0.05$), a more favourable overall assessment was observed for subjects in the HairMax LaserComb® group compared with those in the sham device group.

Safety and Tolerability

The HairMax LaserComb® device was found to be well tolerated. No serious adverse effects were reported. The only adverse events considered to be possibly device related were four cases of mild paraesthesia and four cases of mild urticaria. These showed no statistical difference between study groups. Changes in vital signs from baseline were very small in both treatment groups and similar between both groups.

Discussion

While many unknowns remain, an important component of the treatment of AGA is to provide non-biased demonstration of laser phototherapy effectiveness in hair growth stimulation in humans. Here we report on the first known double-blind, controlled trial of laser phototherapy for the treatment of AGA, that is, sex hormone-dependent male pattern hair loss. Overall, the results of the trial demonstrate significantly greater increase in mean terminal hair density (primary effectiveness) in subjects treated with the HairMax LaserComb® device over the sham device ($p < 0.0001$). Consistent with this evidence for primary effectiveness, significant improvements in overall hair regrowth were demonstrated in terms of patients' subjective assessment ($p < 0.01$) at 26 weeks over baseline. Subjects in the HairMax LaserComb® group perceived significantly greater improvement ($p < 0.05$) regarding overall hair improvement, slowing of hair loss, thicker feeling hair, better scalp health and hair shine.

This study was a pivotol part of the Premarket Notification 510(k) submission in February 2006 and subsequent clearance for marketing by the FDA in January 2007. To date, no other laser therapy device has been cleared by the FDA for marketing and all other similar products on the market are sold as cosmetic devices. This clearance means that the HairMax LaserComb® has been the subject of a rigorous review and clearance process, which differentiates it from other marketed devices that have no clinical proof of efficacy. Thus, it is impossible to compare the HairMax LaserComb® with other devices marketed without clinical studies or FDA clearance.

Conclusions

The current study has accomplished an important goal. This is the first study demonstrating efficacy in hair growth with a laser phototherapy device, the HairMax LaserComb®. This randomized, double-blind, sham device-controlled, multicentre efficacy trial indicates that the HairMax LaserComb® laser phototherapy device with its patented hair-parting teeth mechanism is an effective, well tolerated treatment for hair loss of androgenetic aetiology. Indeed, the HairMax LaserComb® is currently the only laser therapy device that has been clinically studied and proven to grow hair in males with certain classes of AGA.

In the future, the efficacy of HairMax LaserComb® should also be evaluated in subjects with hair loss of non-androgenetic aetiology. It will also be very important to establish the cellular and molecular mechanisms behind the hair growth-promoting effect of laser phototherapy. Future research should help us to differentiate if laser phototherapy predominantly: (i) stimulates anagen re-entry by telogen hair follicles; (ii) increases rates of proliferation in active anagen hair follicles; (iii) prevents premature catagen development; or (iv) extends the duration of the anagen phase. Cellular events, such as activation of dormant hair follicle stem cells, or increase in proliferation of hair matrix trichocytes, should be investigated. In addition, subcellular and molecular signalling events, such as direct short-

term activation of the mitochondrial electron-transport chain, or long-term up-regulation of growth factors, should be evaluated.

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