Clinical & Experimental Ophthalmology

RANZCO The Royal Australian and New Zealand College of Ophthalmologists

Clinical and Experimental Ophthalmology 2014; 42: 466–479 doi: 10.1111/ceo.12247

Original Article

Nanosecond-laser application in intermediate AMD: 12-month results of fundus appearance and macular function

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Abstract

Background: A novel, ultra-low energy nanosecond laser (retinal rejuvenation therapy) has been developed with the aim to slow progression of early age-related macular degeneration (AMD). The safety, changes in fundus characteristics and macular function in a cohort of participants with bilateral intermediate AMD are reported.

Design: Prospective non-randomised, pilot intervention study.

Participants or Samples: Subjects with bilateral intermediate AMD ($n = 50$, aged 50–75 years).

Methods: Ultra-low energy laser pulses applied in 12 spots around the macula of one eye (0.15–0.45 mJ), using 400 μm diameter spot, 3 nanosecond pulse length, 532 nm wavelength and energy titrated to each patient.

Main Outcome Measures: Best corrected visual acuity, drusen area and macular sensitivity (flicker perimetry) at baseline and at 3, 6 and 12 months post-laser.

Results: Treatment was painless with no clinically visible lesions. No participant developed choroidal neovascularization, while two with thin central retinal thickness at baseline developed atrophy at 12-month follow up. Drusen area was reduced in 44% of treated eyes and 22% of untreated fellow eyes, with changes in drusen and function not being coincident. Improvement in flicker threshold within the central 3° was observed in both the treated and untreated fellow eyes at 3 months post-laser. Of the 11 eyes at greatest risk of progression (flicker defect >15 dB), seven improved sufficiently to be taken out of this high-risk category.

Conclusions: A single unilateral application of nanosecond laser to the macula produced bilateral improvements in macula appearance and function. The nanosecond retinal rejuvenation therapy laser warrants ongoing evaluation as an early intervention for AMD.

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Received 16 June 2013; accepted 16 September 2013.

Competing/conflicts of interest: Ellex provided the laser and contributed some funds to CERA to support the pilot study. Ellex's participation related to protocol development and advice regarding the evaluation of the visual function results. AV provides consultancy to Medmont Pty Ltd and receives research support from the company.

Funding sources: This work was supported by a grant to the Centre for Eye Research Australia, from the Victorian State Government under the Victoria's Science Agenda Investment Fund and from Ellex R&D Pty Ltd, Adelaide. RHG receives a National Health and Medical Research Council (NHMRC) practitioner fellowship (#529905). CERA is supported by a NHMRC Centre for Clinical Research Excellence #529923 award and receives operational infrastructure from the Victorian Government. The funding organizations had no role in the design or conduct of this research.

Key words: AMD, macula, nanosecond laser, perimetry.

INTRODUCTION

Early signs of age-related macular degeneration (AMD) such as drusen and pigmentary abnormalities can be diagnosed through a simple eye examination, often years before sight-threatening complications occur. While current advice to slow progression centres around changes in lifestyle and diet, there is no specifically targeted intervention to offer.[1–3] There are often many years in which to intervene with an early intervention strategy if one existed.

A novel, ultra-low energy nanosecond (ns) laser, retinal rejuvenation therapy (2RT), has been developed by Ellex R&D Pty Ltd (Adelaide, SA, Australia) with the aim of slowing progression of early AMD. Using very short laser pulses (3 ns), an insult caused by steam production around melanosomes can be confined to the retinal pigment epithelium (RPE), inducing a highly selective and discrete non-thermal injury.[4–8] While the mechanism by which the insult to RPE cells brings about a beneficial change in the macula is not well understood, it has been hypothesized that a 3-ns laser could induce migration of RPE cells and release of matrix metalloproteinases, improving the hydraulic conductivity of Bruch's membrane.[9,10] Whatever mechanisms are at play, it is clear that any beneficial effects to the macula occur without the potential harmful effects seen with traditional thermal lasers, thereby offering a new opportunity to consider it as a prophylactic treatment of early AMD.[7,8]

With any new intervention for early AMD, a significant impediment to its implementation is the lack of an outcome measure that can be monitored over time to determine if there is a slowing in progression or reversal of early AMD changes. Although assessment of structural changes has improved in recent years, through methods such as optical coherence tomography (OCT) and fundus autofluorescence (FAF),[11,12] these methods lack a functional component. The traditional approach to assessing retinal function is with visual acuity (VA). However, in early AMD, acuity is often within the normal range.[13] Better-targeted functional tests are needed to monitor progression and assess novel interventions that aim to slow progression towards vision loss.

Previous studies have indicated a broad range of functional abnormalities in eyes with early stages of AMD.[14–28] In several studies, including our own, flicker sensitivities, recorded with perimetry revealed losses and appeared to be a clinically applicable test to follow changes in retinal sensitivity over time.[17,23–28] We have recently shown that flicker sensitivities appeared to predict eyes at risk of developing advanced AMD. Eyes that went on to develop geographic atrophy (GA) or choroidal neovascularization (CNV) had a significantly reduced mean flicker sensitivity in the months prior to the development of late stage disease.[28] We therefore investigated the ability of functional testing – flicker perimetry – to follow participants in this intervention study to determine if there was evidence of functional change at the macula.

Our aim was to conduct a pilot study of the 2RT laser in participants with bilateral intermediate AMD, at high risk of developing sight-threatening complications of AMD,[29] to investigate if this treatment could alter the clinical signs of the early stages of AMD and to improve or stabilize retinal function, as determined by flicker sensitivities.

METHODS

The study was approved by the Human Ethics Committee of the Royal Victorian Eye and Ear Hospital and research adhered to the tenets of the Declaration of Helsinki. This trial has been registered with the Australian New Zealand Clinical Trials Registry. Trial ID: ACTRN12609001056280.

Study participants

Participants with bilateral intermediate AMD (drusen >125 μm)[30] were recruited to take part in the pilot study of 2RT laser. Participants were recruited from the medical retinal clinics at the Royal Victorian Eye and Ear Hospital or were referred by retinal consultants.

Main inclusion criteria were age greater than 49 years, with bilateral, intermediate AMD (multiple drusen >125 μm in both maculae), a best corrected visual acuity (BCVA) using standard Early Treatment Diabetic Retinopathy Study logMAR chart of 6/18 (60 letters) or better and willingness to sign an informed consent. Main exclusion criteria were any evidence of GA on colour fundus photographic grading, presence of CNV, any past treatment for CNV in either eye or signs of any other ocular disease that would prevent follow up evaluation. Follow-up visits were at 3, 6 and 12 months.

Control population database for flicker perimetry analysis

While the perimeter used measures of absolute sensitivity levels between 0 dB (no response) and 45 dB (maximum sensitivity), with a typical normal value of 25 dB, the graphs within this paper show deviation from age-matched normal averages.

Our own database of 72 flicker perimetry results from 30 normal participants without AMD, ranging

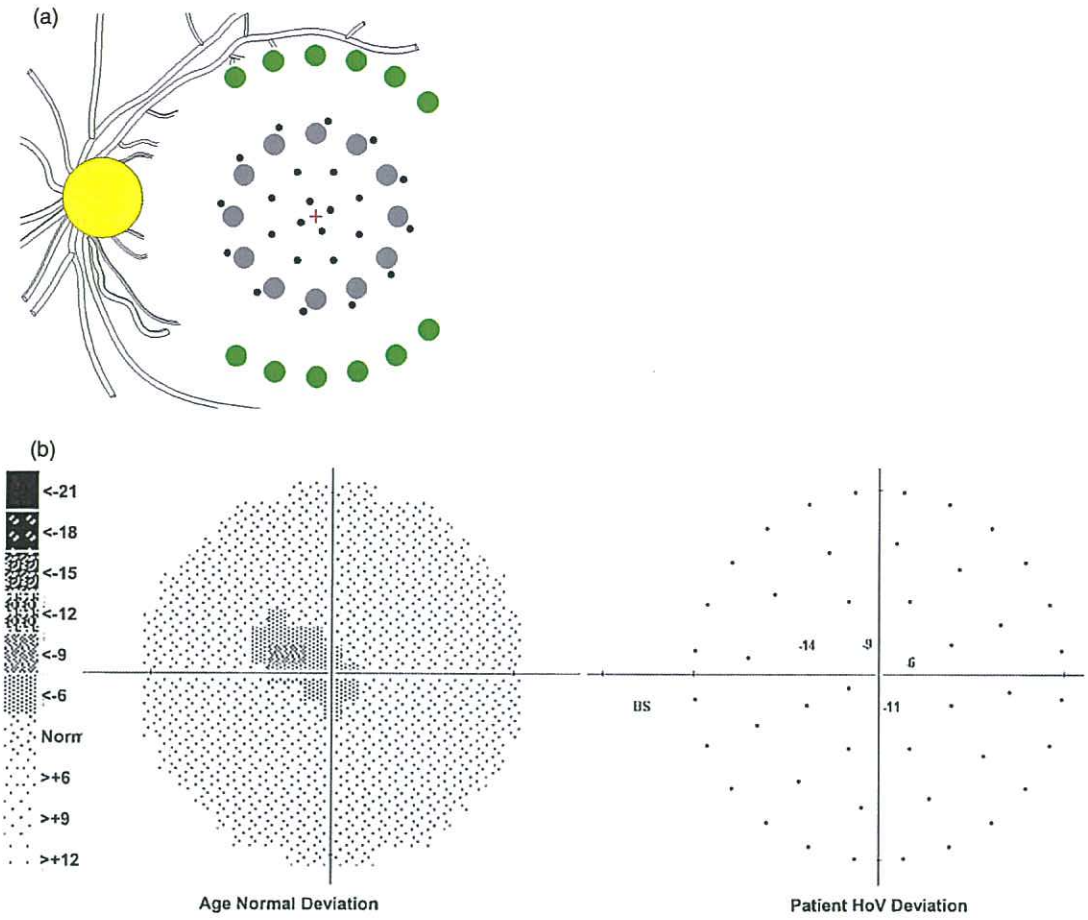

Figure 1. (a) Position of the central 24 perimetry test points (black dots) relative to visual angle. Mid macula (grey spots) laser treatment locations for Protocol 1 and outer macula (green spots) laser treatment locations for Protocol 2. Red cross indicates foveal centre. (b) An example of the Medmont perimeter output in early age-related macular degeneration (AMD). Graphical representation (left), and deviation in dB, from age corrected normal (right). HoV, hill of vision.

in age from 16 years to 85 years old, was used to age correct all visual field tests results. Of these, there were 23 control participants, over 49 years of age, with the same inclusion and exclusion criteria. These yielded age-related trend lines for the average of all points and also the 1, 3 and 6 degree rings. Formulae of these trend lines were used to produce age corrected deviations from average, for each trial participant. Average normal values for worst single point defect and 5-point cluster defect were also determined from the same normal database.

Investigations performed

An AMD risk factor questionnaire, BCVA, ophthalmic examination, colour fundus photography (Canon CR6-45NM Non-Mydriatic Retinal Camera, Tokyo, Japan), auto-fluorescence images (Spectralis HRA+ OCT; Heidelberg Engineering, Heidelberg, Germany) infrared (IR) reflectance images (Spectralis HRA+ OCT) and OCT scans of the macula (Spectralis HRA+ OCT and Cirrus HD-OCT; Carl Zeiss Meditec, Inc., Dublin, CA, USA) were performed pre-treatment and at each follow up. Macular flicker perimetry tests (model M-700; Medmont International Pty Ltd, Nunawading, VIC, Australia) were obtained on all participants as a clinically applicable psychophysical macular function test.[16,26–28]

Flicker perimetry

Flicker perimetry was performed using methods previously reported.[25] In brief, the M-700 is a bowl perimeter using light-emitting diodes [565 nm light emitting diodes (LEDs)], with background luminance of 3.2 cd/m^2 and maximum spot luminance of 320 cd/m^2. The LEDs subtend 0.43° (Goldmann III) and are arranged concentrically at various eccentricities, from 1 to 50 degrees. We used the macular test protocol and report on the 1, 3 and 6 degree rings (Fig. 1a,b). The test took between

4 and 7 min to complete per eye. Flicker thresholds were determined using the autoflicker test where thresholds were obtained at 1° and 3° with stimuli flickering at 18 Hz, 16 Hz at 6° and 12 Hz at 10°.[31] This method has been used successfully in the clinical settings of migraine, diabetes and glaucoma.[32,33] We recorded the flicker perimetry twice, with an average being taken for baseline, then 3, 6 and 12 months post-laser.

A natural history perimetry cohort[26] with similar bilateral intermediate AMD changes and VA was used to determine test re-test reproducibility. Using 41 participants with two tests 6 months apart, we found the test re-test reproducibility had a standard deviation of 2.34 dB for flicker perimetry with 95% (2 standard deviation) of values falling within 4.68 dB.

AMD diagnosis and grading

At screening a dilated fundoscopic examination was followed by digital fundus photography. Determination of inclusion criteria and eligibility for the study was based upon the clinical examination of the retina and grading of colour retinal images. Participants with any evidence of GA or CNV seen clinically or on colour photography were excluded from the study. When indicated, fluorescein angiography was undertaken to verify the absence of CNV.

Subsequently all images, including the FAF and OCT images, from baseline to 12 months, were graded for the presence of GA or CNV. The baseline and 12 months images were graded by two graders, who were masked to the treated eye and whether the photos were baseline or 12 months, for drusen area as well as side by side grading for 'same, better, worse' in terms of drusen area. Grading of drusen area was (changes ± 5% were clinically irrelevant) compared with the side by side grading. Change in pigment was not used as a marker of disease severity as the laser treatment occasionally created pigmentary changes at the site of the treatment. Any inconsistencies in grading were resolved by two other clinical investigators.

Drusen volume within the central 3 mm ring was obtained in a subgroup (SG) of subjects ($n = 17$) using the macular cube scans of the Cirrus HD-OCT instrument (Carl Zeiss Meditec, Inc.). This was because the equipment was not available at the commencement of the study. The analysis function available within the Cirrus system automatically generated a report of drusen volume estimation. This algorithm for drusen volume estimation has been validated previously.[34,35] The change in drusen volume between baseline and 1-year post-laser was calculated.

Nanosecond laser

The 2RT laser delivers single pulses at a wavelength of 532 nm with a pulse duration of 3 ns. Laser spot size is fixed at 400 μm diameter, with a fine speckle energy distribution beam profile applied coaxially through a slit-lamp microscope and 1:1 macular contact lens (Area Centralis Volk Ophthalmic Inc., Mentor, OH, USA). Test shots were placed inferiorly outside the arcades to determine the visual effect threshold (retinal blanching just visible) and, once determined, laser energy was reduced by 20% and applied as the treatment to the macula. The laser was not intentionally directed towards drusen, but rather just evenly distributed at a safe distance from the fovea. Twelve spots were initially placed in a clock face pattern 5° radius (1500 μm) from the foveal centre (protocol 1) (Fig. 1a), using the individualized energy level. With experience, we found that the effect was widespread, and given that this study was an exploratory, first in AMD pilot study, we changed the protocol so that the treatment spots were moved out slightly further from the foveal centre (approximately 2000 μm), to just inside the arcades (protocol 2) (Fig. 1a). The treatment energy was approximately 1000 times less than is used in thermal laser treatment to the macula for diabetic macular oedema and uses pulses approximately 33 000 000 times shorter in duration. A maximum limit to laser intensity was set at 0.3 mJ after one participant developed a dot haemorrhage after a test shot at 0.45 mJ, although no visual effect threshold was reached. The laser was applied only to the worst performing eye based on BCVA, then flicker sensitivities if BCVA was the same, and if both eyes were equal than the eye, the patient perceived as their worst eye.

Flicker perimetry analysis

We were particularly interested in the worst performing test point in each of the central three rings of different eccentricities (1, 3 and 6 degrees) (Fig. 1a) as sensitivity losses in early stages of AMD are usually limited to a small region and hence averaging points is not useful. Our analysis was limited to the central 6°.[26] As a mean of verifying the single point analyses, the same analysis was performed using a 5-point cluster defect, averaging over a region that included the four neighbouring points as well as the worst single point (results not shown).

High-risk SG

Many participants had normal pre-treatment sensitivities so that it was not possible to determine improvement in sensitivity. We therefore analysed separately a smaller SG of patients who had

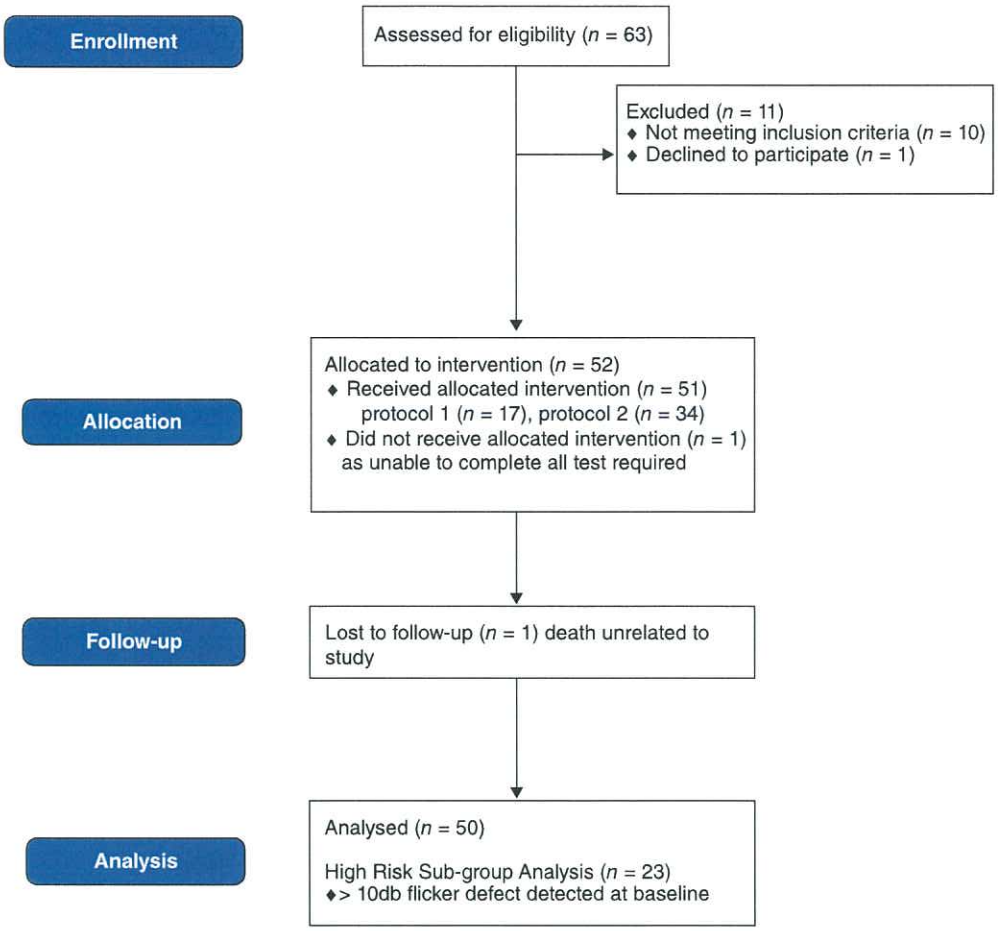

Figure 2. Flow chart of participant recruitment.

abnormal baseline perimetry results, as improvements were possible in these eyes. For this SG, we selected participants who had a worst point of >10 dB deviation in either eye (from aged-matched controls), who we considered at highest risk of development of GA.[28] We excluded from this SG three patients who on FAF had areas of hypo-autofluorescence that were determined to be areas of atrophy, but which had not been detected on the baseline colour fundus images or on clinical examination. Twenty-three participants were included in this separate high-risk SG as well as being included in the analysis of the entire cohort.

Results

Baseline characteristics

Sixty-three people were screened, with 10 not meeting the inclusion criteria and 1 declining to participate. Fifty-two participants with bilateral intermediate AMD were enrolled with the average age being 68 years (49–86 years). Thirty-six patients were female and 16 male. BCVA ranged from 93 Early Treatment Diabetic Retinopathy Study letters (6/4.8) to 60 letters (6/18). One participant, unable to perform the baseline tests, was exited before treatment, and one participant died aged 78 of an unrelated illness within 2 months of treatment and so was not included in anything other than the baseline analysis. Therefore, 12-month follow-up results are provided for 50 participants in this paper (Fig. 2).

On review of all baseline images, three participants had evidence of atrophy that was not recognized clinically or on colour fundus images. Two had thin central retinae on the OCT thickness map, and one had an unusual appearing atrophic extrafoveal area. These three were included in the overall analysis but excluded from the high-risk SG as they, in retrospect, already had signs of advanced AMD (GA) when all imaging modalities were considered.

Baseline flicker perimetry

The average pre-treatment worst point defects in flicker sensitivity by visual angle are plotted in

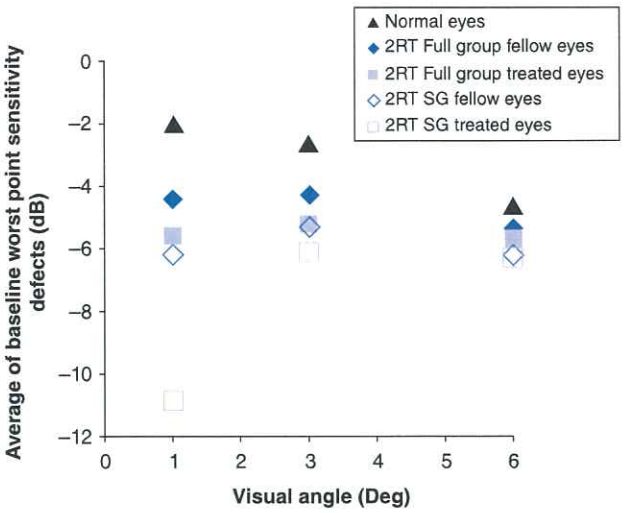

Figure 3. Average pre-treatment worst point visual sensitivity defects by visual angle, measured using flicker perimetry for age-matched controls (black triangles), retinal rejuvenation therapy (2RT) full group of treated eyes (solid squares) and full group of fellow eyes (solid diamond), the treated eyes of the high-risk subgroup (SG) (hollow squares) and fellow eye of high risk subgroup (SG) (hollow diamonds), The *Y* axis zero line is the average of all points in normal eyes. Note that the greatest defect was seen in the eyes selected for treatment and in the para-foveal 1° region.

Figure 3. The average of the worst point sensitivity defect in the 1° ring of the normal cohort was about 2 dB indicating that points in normal maculae can show small levels of variability in sensitivity whereas the greatest reductions seen in the AMD group were between 4 and 11 dB (Fig. 3). The worst flicker sensitivity occurred in the eye selected for treatment, in line with the treatment protocol, and the 1° and 3° rings of the high-risk SG eyes selected for treatment had worst point average visual function compared with all other groups.

Nanosecond laser treatment

Fifty-one participants were treated in one eye with ultra-low energy laser pulses (2RT laser) applied in 12 spots around the macula, titrated to suit each patient. The average laser energy at each treatment spot was 0.24 mJ (with a range of 0.15–0.45 mJ) with an average radiant exposure of 0.19 J/cm^2 (ranged 0.12–0.36). One patient developed a dot haemorrhage during the test shots when 0.45 mJ was applied. The laser procedure was abandoned, and no macular laser was ever given. We continued to follow this participant who became one our best responders over the 12-month period. After this participant, we limited the maximal laser energy to 0.3 mJ, and protocol 2 was adapted for the remaining 34 participants (Fig. 1a). We initially performed separate analyses for each protocol and found that all outcome parameters were of similar trends irrespective of the protocol used. We therefore combined the data from both protocols for the final analyses.

Laser spots were not seen on colour fundus photos, IR reflectance or FAF imaging immediately after the laser; however, after 3 months, they could usually be seen on IR reflectance or FAF images, but not standard photography and appeared as areas containing small speckles which faded over time (Fig. 4). On OCT, they could occasionally be seen as a discrete increase in signal at the level of the RPE, corresponding to the areas where a change was seen on the IR reflectance and FAF images (Fig. 4b). No evidence of photoreceptor or inner retinal damage on OCT was seen. Treatment was painless and easily tolerated with some participants reporting an after image of the spots lasting from minutes to a few days. No participant developed CNV. We also observed a similar effect in the untreated fellow eyes, in both drusen resolution and improved retinal function. This was unexpected and rendering the untreated fellow eye unable to be considered as the control eye.

Individual patient examples

Examples of individual patient responses are provided in Figures 5–7 to give an overview of the range of changes seen.

Visual acuity

Overall, the average BCVA did not change at 12 months post-treatment (−0.1 Early Treatment Diabetic Retinopathy Study letters in treated eyes and 0.8 letters in non-treated fellow eyes) (Fig. 8). Of the 50 treated eyes, eight improved by five or more letters, and seven lost five or more letters over 12-month follow up. In the untreated fellow eyes, four improved five or more letters, and four lost five or more letters over 12-month follow up. Of those that lost five or more letters in the treated eyes, two had thin central retinal thickness on OCT at baseline with one also having a subfoveal pigment epithelial detachment (PED) >1000 μm at baseline. These two proceeded to develop clinically apparent GA over the 12-month follow up. Another participant developed a clinically significant cataract and another had a large PED >1000 μm at baseline. For the other three participants, there was no obvious explanation for the drop in BCVA.

Change in drusen area and volume

Reduction in drusen area was found in 44% of treated eyes over 12 months, with 24% increasing in

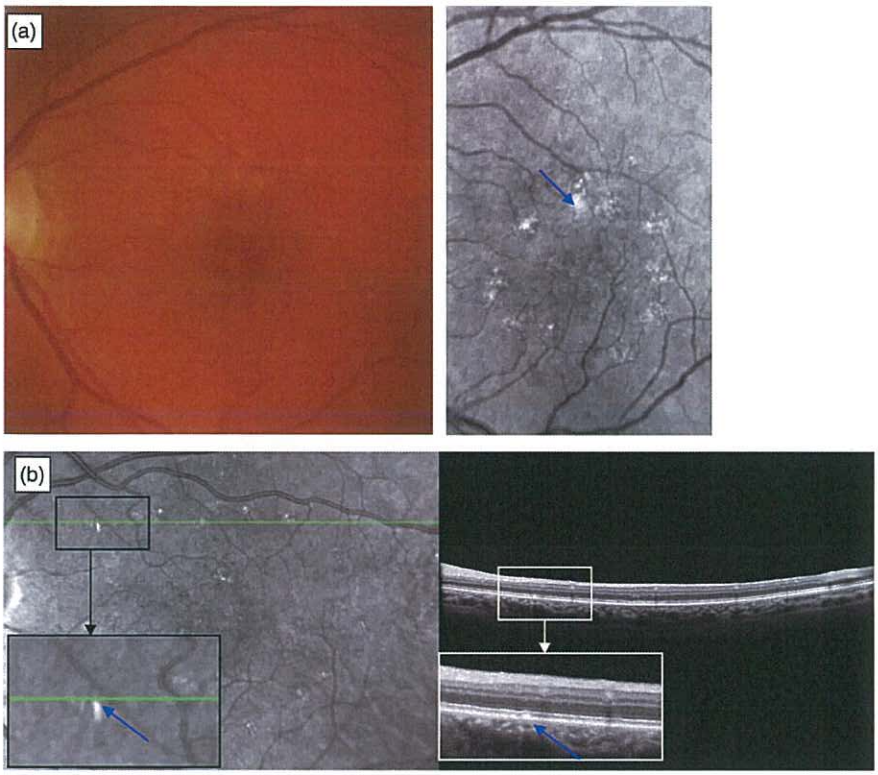

Figure 4. (a) Fundus photo (left) and IR reflectance image (right) of a 67-year-old patient at 3 months (blue arrow on IR reflectance image indicates laser the spot). (b) An optical coherence tomography (OCT) image of a 71-year-old participant showing the laser spot 12 months after the treatment. The retinal pigment epithelium (RPE) shows an increased signal but there is no other obvious change throughout the retina. Blue arrows mark the same hyper-reflective spot on both images.

area. In the untreated fellow eye, 22% showed reduced drusen area at 12 months while 18% increased in area (Fig. 9). When there was a reduction in drusen there was often a reduction in the hyper-autofluorescence in the same area on the FAF image (Fig. 6).

Of the 17 subjects who had Cirrus HD-OCT scans performed at baseline and at 12-month post-laser, seven (41%) participants had a reduction in drusen volume of at least 0.028 mm^3 in the treated eyes at 12 months.

Visual function changes

In many patients, reduced flicker sensitivities was often limited to a small (often foveal-perifoveal) area, with relatively normal function in other areas. We therefore present the worst point analysis of flicker sensitivity over time for the treated and untreated fellow eyes (Fig. 10). In ring 1 (1°), the sensitivity of the treated eyes in both the full group and high-risk SG improved, on average, by at least 9 dB at 3 months post-laser. This was the region of greatest pre-treatment defect (Fig. 3). This improvement was greatest at 3 months, reducing at 6 and 12 months post-laser; however, on average, the sensitivity at 12 months remained greater than the baseline values. The effect of the 2RT laser on the flicker sensitivity of the fellow eye was minimal within the central 1° retina. In ring 2 (3°), however, the sensitivity of the fellow eye gradually improved over time while there was no improvement in retinal function detected in the treated eyes. In ring 3 (6°), where there was little difference in sensitivity compared with age-matched normal at baseline, there was no improvement in the flicker sensitivity of either the treated or untreated fellow eyes at all time points post-laser.

Individual results of the entire 23 high-risk SG are shown in the scatter graph (Fig. 11) indicating the wide range of changes to worst point sensitivity after treatment to the study eye. Patients with little pre-treatment defect (top right corner) show little or no improvement; however, the majority of those with large pre-treatment defects show large improvements (above the 'no change' line) at 6 and 12 months post-treatment. The change evident at 6 months is generally sustained at 12 months.

We found a close match between 5-point cluster results and single worse points (results not shown) indicating that regions larger than a single point were improving, although the average of all the tests points remained stable at a near normal level suggesting that no learning effect was present in the data.

We have previously reported that people who have sensitivity defects of greater than 15 dB compared with age-matched controls are at very high risk of progressing to advanced disease.[28] We therefore determined what percent of people we were able to 'rescue' from this position. Figure 12 represents the percent of

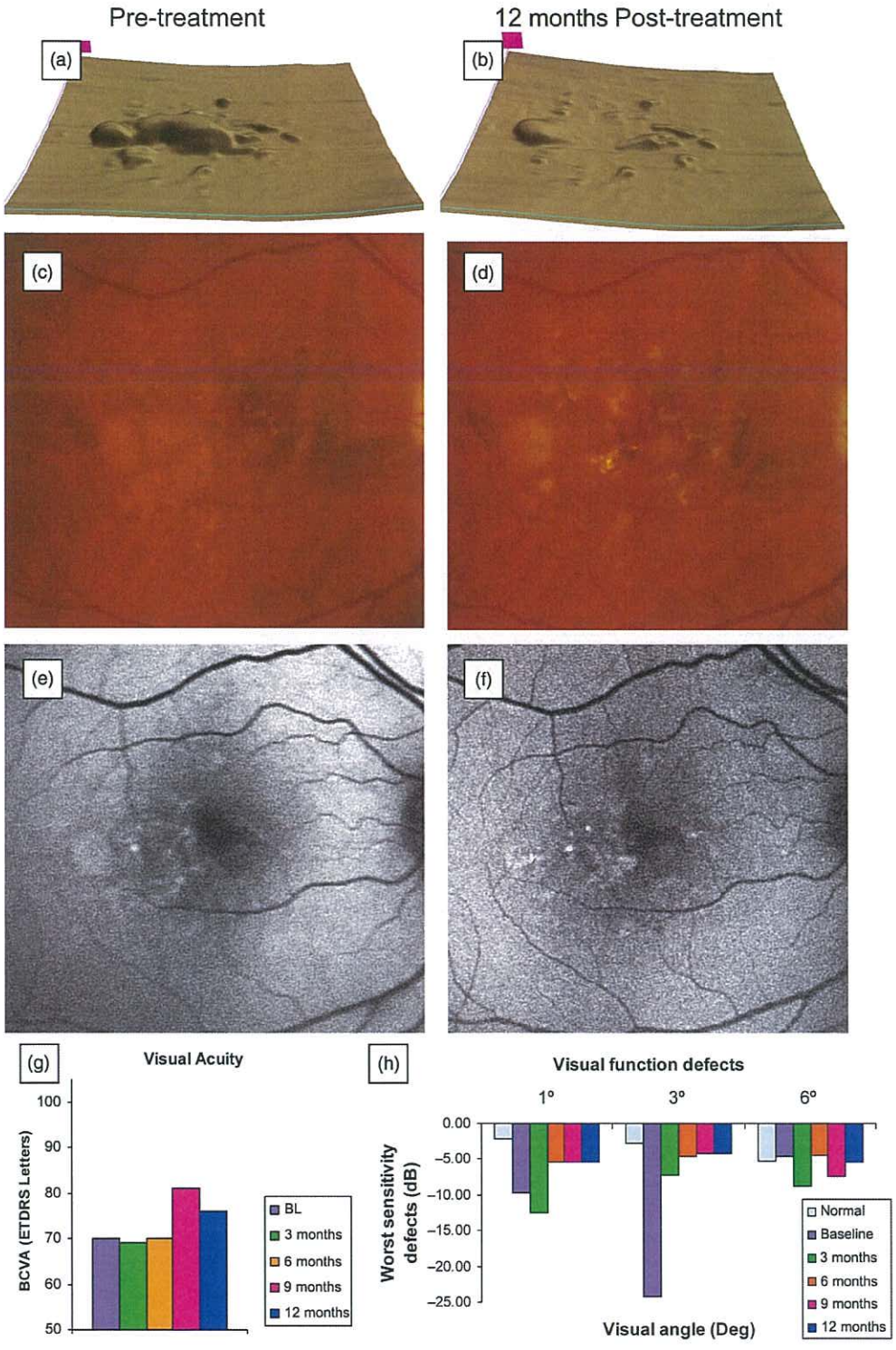

Figure 5. Pre-treatment (left side) and 12-month post-treatment (right side) physical and function changes in the treated eye (OD) in a 72-year-old participant using laser protocol 1. From top (a,b) retinal pigment epithelium (RPE) layer maps from spectral domain optical coherence tomography (OCT), (c,d) fundus photos, (e,f) auto-fluorescence (FAF) images, (g) visual acuity (VA) changes and (h) visual function changes at the points of worse visual sensitivity defect. Note the reduction in para-foveal drusen and hyper-fluorescence on the FAF image and the improvement in both VA and flicker sensitivity, especially at the worst location of the 3° ring.

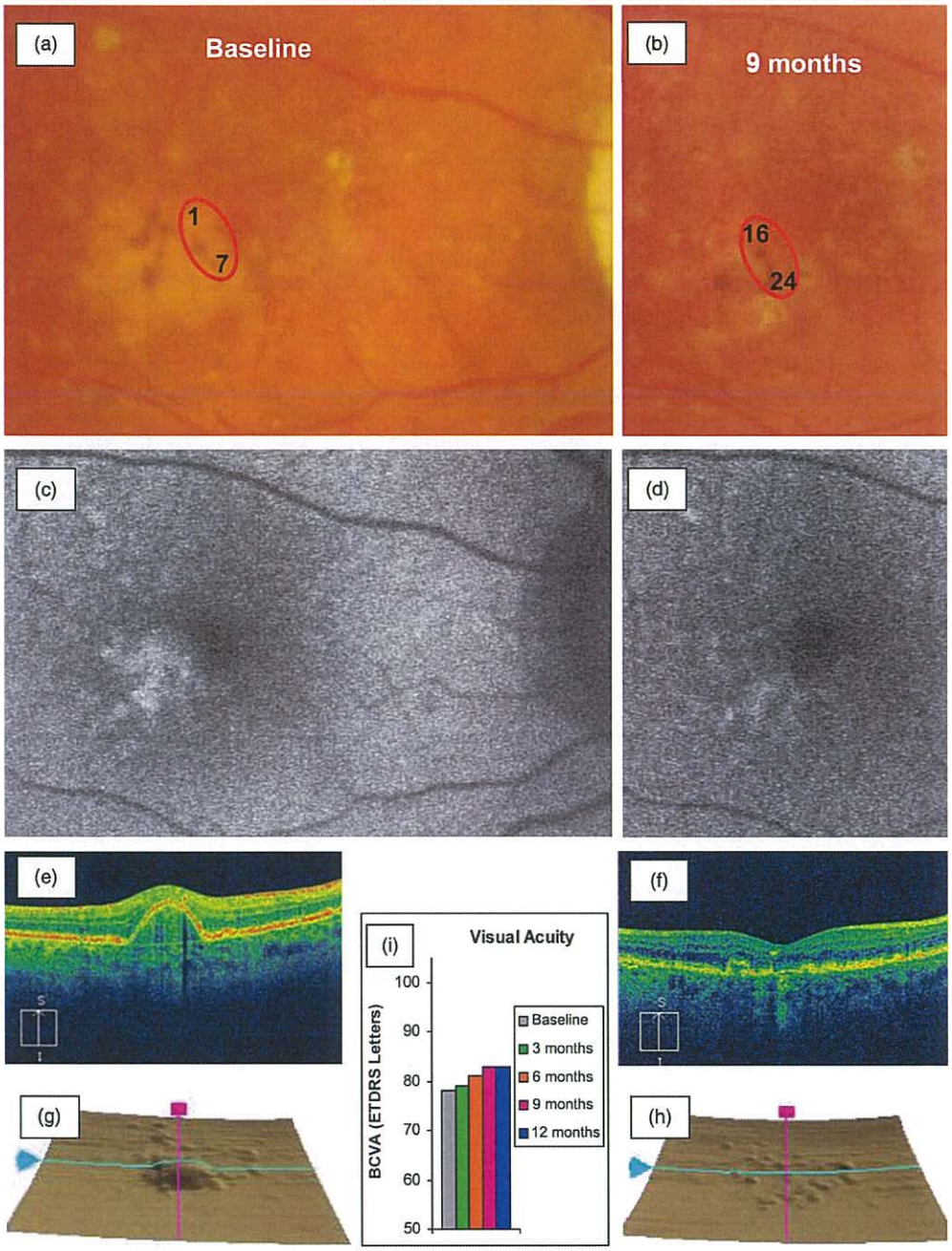

Figure 6. Functional and physical changes in the treated eye (OD) of a 73-year-old participant before and 9 months after retinal rejuvenation therapy (2RT) laser (Protocol 2). (a) Pre-treatment macula photo with measured absolute perimeter values of the major visual sensitivity defect (red oval) in approximate locations in the 1° ring. All other points were within the normal age matched limit. (b) 9 months photo showing large drusen reduction and movement of pigment, along with improved perimeter values. (c) Pre-treatment auto-fluorescence (FAF) image showing hyper-fluorescent regions within drusen. (d) 9 months FAF image showing large reduction in hyper-fluorescence. (e) Pre-treatment optical coherence tomography (OCT) showing para-foveal PED. (f) 9 months OCT showing PED resolution. (g and h) Pre-treatment and 9 month retinal pigment epithelium (RPE) layer map from OCT images e and f. (i) best corrected visual acuity (BCVA) showing gradual improvement over 12 months.

eyes where at least one point returned flicker sensitivities worse than 15 dB below age-matched normal at baseline, in the full group ($n = 50$) and the SG ($n = 23$). In the high risk SG, 11 (48%) eyes were in this category. Of these, after treatment, seven (64%) were 'rescued' from this very high-risk zone at 6 months. The reduction seen in the high risk SG was significant at 6 months ($P = 0.027$).

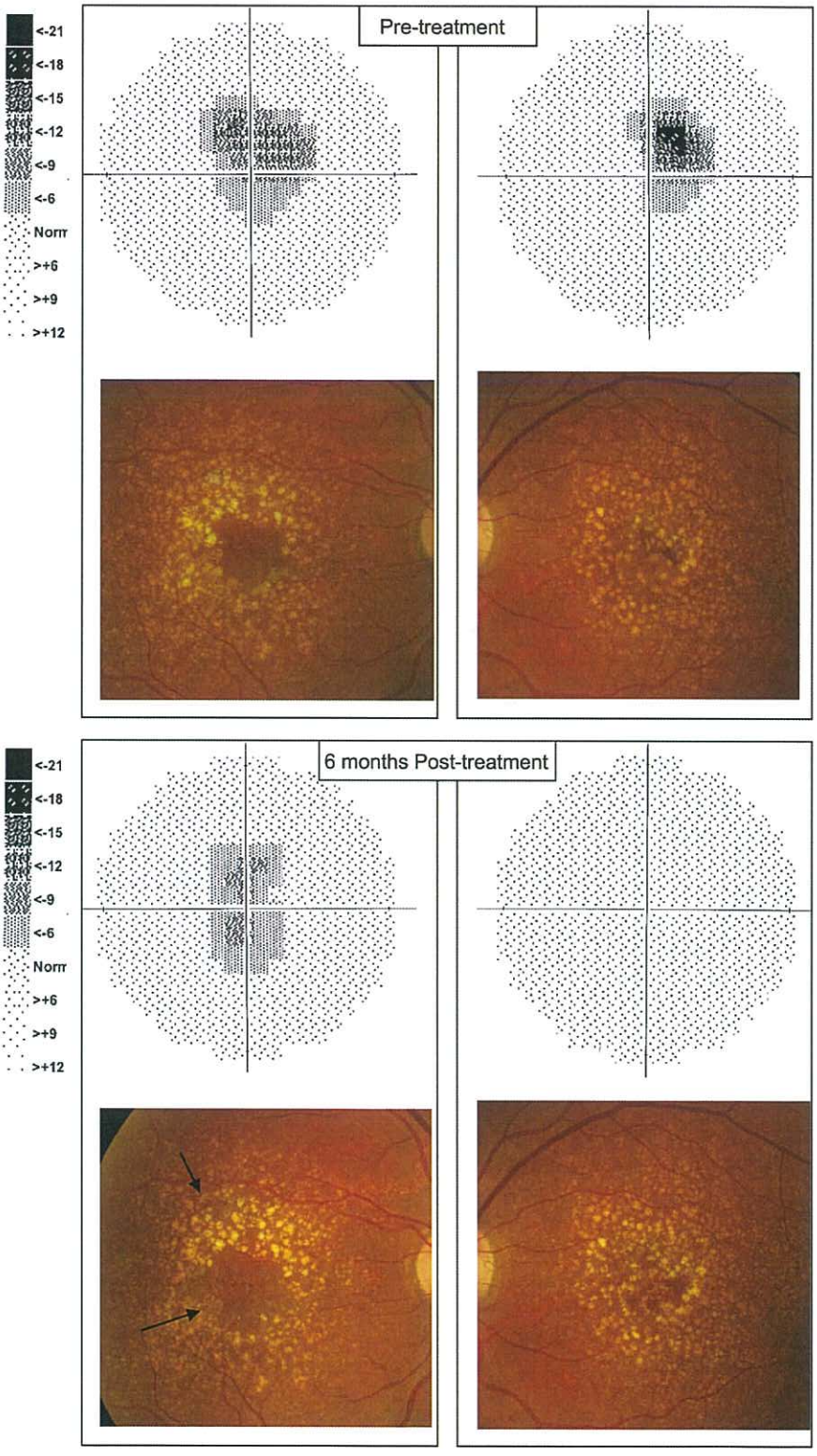

Figure 7. Graphical representation of flicker perimeter deviations from age-matched normal and macular photos of both eyes of a participant aged 55 years, before (top) and 6 months after (bottom) laser treatment (Protocol 2) to right eye. Black arrows show drusen reduction in the treated eye and improved visual sensitivity defect in fellow eye. While there was some improvement in the visual field defect in the treated eye, the defect was completely reversed in the non-treated eye and flicker sensitivity restored to within the age-matched normal range. It can also be seen that functional defects were not coincident with drusen location nor were regions of functional improvement coincident with drusen resolution.

Discussion

In AMD, we lack an intervention specifically aimed at slowing or reversing the early changes seen in AMD that indicate risk of progression to advanced AMD and vision loss. We present here the first results from a novel intervention with a nanosecond laser aimed at slowing progression of the early stages

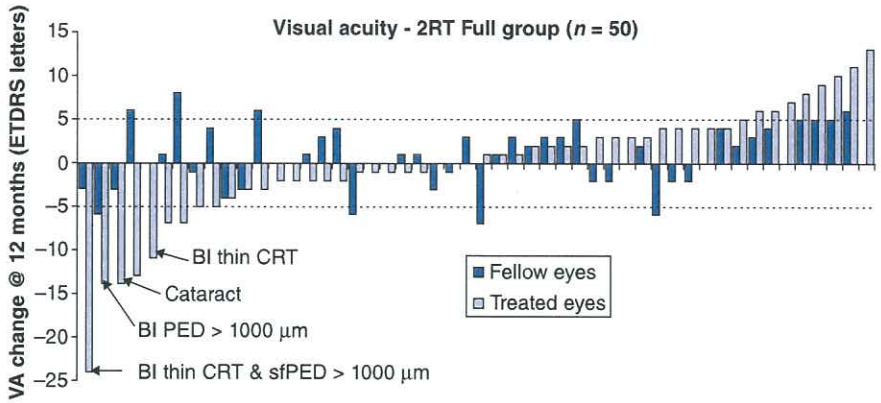

Figure 8. Changes in best corrected visual acuity (BCVA) over 12 month follow up, in treated eyes (light blue) and fellow eyes (dark blue) of retinal rejuvenation therapy (2RT) full group (n = 50). ±5 letters shown as dashed lines. Each participant was ordered according to the change in the treated eye. There were examples where the untreated fellow eye improved while the treated eye declined and examples of the reverse. Characteristics thought to be related to VA loss are indicated for the treated eyes that lost more than seven letters. BI, baseline; PED, pigment epithelial detachment; CRT, central retinal thickness.

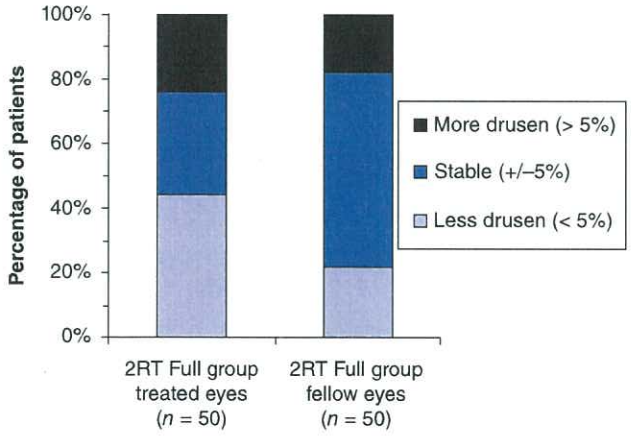

Figure 9. Change in drusen area grading in the retinal rejuvenation therapy (2RT) full group at 12 months, in treated eyes and their fellow eyes.

of AMD. The 2RT nanosecond laser used in this study delivered a radiant exposure over 1000 times less than conventional macular thermal lasers, and produced no clinically visible lesions.

A single treatment resulted in drusen area reduction in 44% of treated eyes. When we used automated drusen volume calculations (41%), participants had a reduction in drusen volume of at least 0.028 mm^3 in the treated eyes. The proportion of subjects with drusen volume reduction (41%) in our study was significantly higher than the value reported in the natural history study[34] of 20% (chi-square = 3.97, P = 0.0464). Over the 12 months of follow up, none of these participants developed GA, which is usually the sequelae of drusen resolution.

This single treatment also improved visual function, as measured by flicker sensitivity, in both the treated and untreated eyes for at least 12 months. Improvements in retinal function in early AMD lends support to the notion that degenerative changes occurring at the level of the photoreceptor/RPE/Bruch's membrane interface may be able to be reversed by treatment, if given early enough. Recently, the 2RT laser has also been shown to be safe and have similar efficacy to conventional thermal photocoagulation when used in diabetic macular oedema.[36]

Bilateral effects from a unilateral treatment have been identified in the use of other nanosecond pulse lasers such as with selective laser trabeculoplasty in the treatment of open-angle glaucoma. Rhodes *et al.*[37] reported an 18.8% reduction in intra-ocular pressure at 6 months post-treatment in the treated eye and an 11.2% reduction in the untreated fellow eye.

Immune privilege in mice has been disrupted in both the eye that had the RPE cells damaged by retinal laser burns as well as the non-treated eye.[38] It has been postulated that the neuronal signals generated from damaged RPE cells of the treated eye had a modifying influence on the contralateral eye as part of an autonomous response that could possibly be nerve mediated. Forrester's group suggest that in AMD, the balance between stress-induced damage and para-inflammation-related tissue repair and remodelling is disturbed.[39] It may be that the laser response changes the dynamics between protective repair mechanisms and the detrimental chronic inflammation in AMD as the result of some systemic mechanism, possibly mediated by circulating factors or immune modulation. Further research is being undertaken in an effort to answer these questions.

Point-wise analysis of the flicker perimetry enabled us to observe changes in the points and

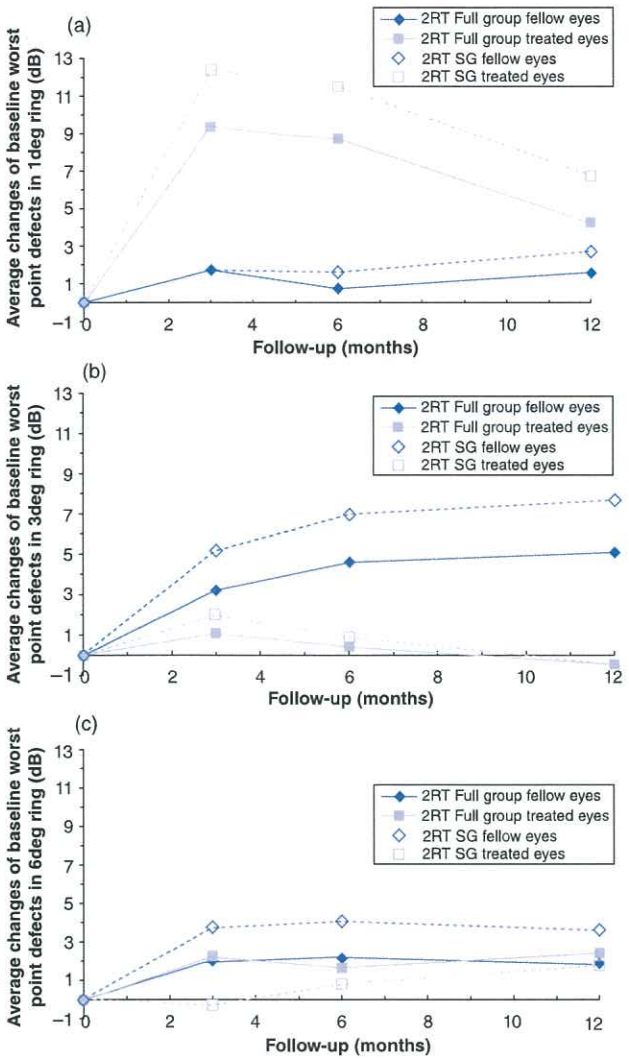

Figure 10. Improvements in flicker sensitivity in worst visual sensitivity defects at a: 1°, b: 3° and c: 6° according to months post-treatment. Solid squares: retinal rejuvenation therapy (2RT) full group treated eye, solid diamonds: 2RT full group fellow eyes, hollow squares: 2RT subgroup (SG) treated eyes, hollow diamonds: 2RT SG fellow eyes. The biggest changes occurred in the central 1° of the treated eye, where greatest pre-treatment dysfunction existed. The greatest effect in the 3° ring was seen in the fellow untreated eye. In the treated eye, the improvement was maximal between 3 and 6 months with a gradual decline after 6 months but not back to pre-treatment levels. Error bars were not presented because the difficulty in their interpretation when using log units.

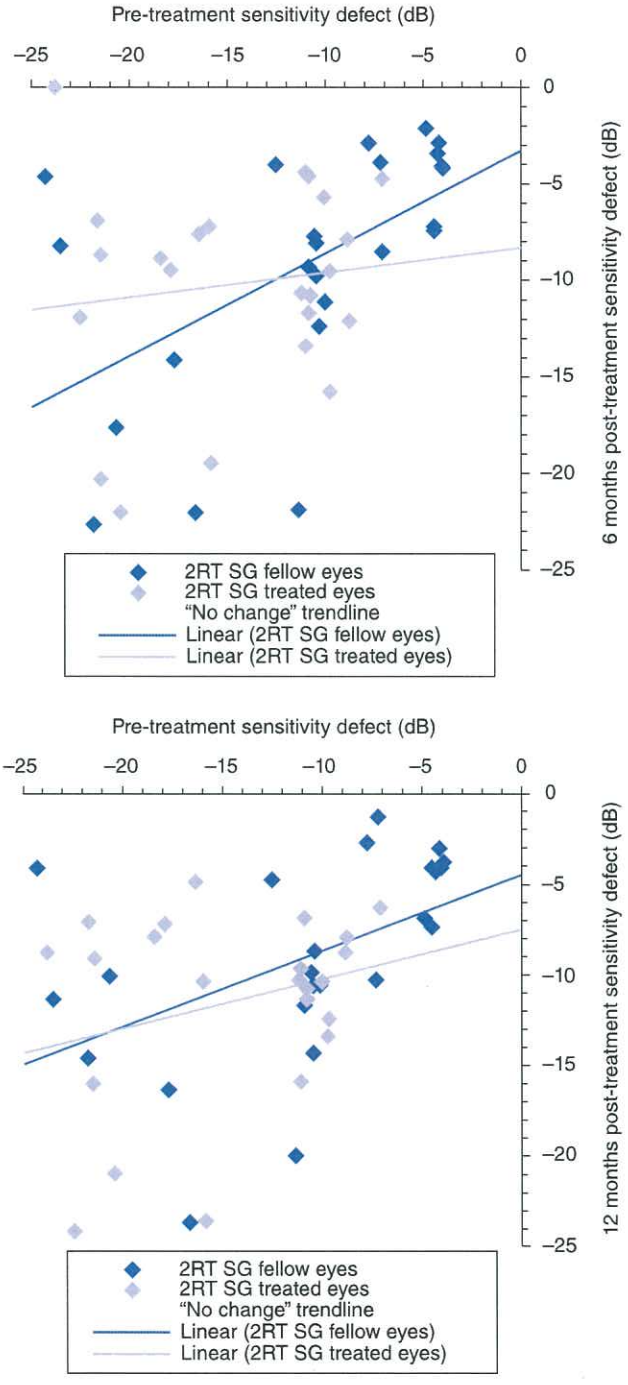

Figure 11. Scatter graphs comparing pre-treatment worst visual sensitivity defects to same point at 6 month (a) and 12 months (b) results of all patients in the retinal rejuvenation therapy (2RT) subgroup (SG) (n = 23). Dark blue diamonds: 2RT SG treated eye, light blue diamonds: 2RT SG fellow eyes with linear trendline shown as solid lines for each group. Dashed line represents a 'no change' result (above this line indicates improvement, below indicates worsening). Patients with little pre-treatment defect (top right corner) show little or no improvement; however, the majority of those with large pre-treatment defects show large improvements at 6 and 12 months post-treatment.

clusters over time. We hypothesize that the worst performing points represent areas most likely to develop atrophy.[28] In our study, we took care to minimize the learning effect by repeat baseline testing in the 2RT-treated group; however, it is likely that a learning component exists within the post-treatment improvements reported in this study. Most patients

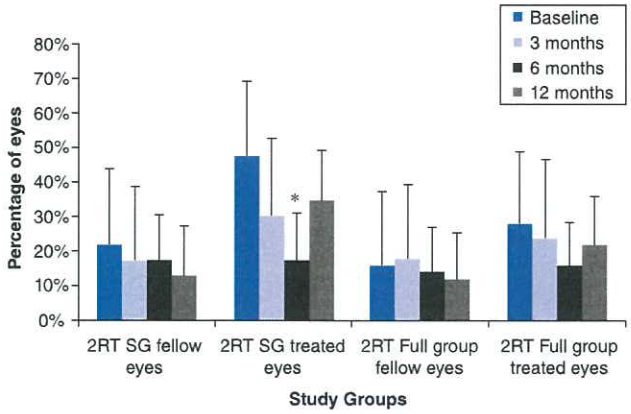

Figure 12. Percentage of eyes where at least one point returned flicker sensitivities worse than 15 dB below age-matched normal in the different study groups at various time points in the full group ($n = 50$) and the subgroup (SG) ($n = 23$). Error bars represent 95% confidence interval. *The reduction seen in the high-risk SG was significant at 6 months ($P = 0.027$).

gave similar sensitivity results across most data points over several time points suggesting that the learning effect in this cohort was minimal.

The improvements seen in this report were largely in the worst performing areas of either eye – the points at greatest risk of progressing to GA and vision loss. As shown in Figure 10, the trends of visual function change suggest that improvements in the treated eye can occur soon after treatment, but the process continues at least for 6 months, with improvement starting to wane but still effective out to 12 months. This suggests that retinal deterioration is not only being halted, but to some degree reversed. Interestingly, Figure 10 shows that the treated eye response peaks between 3 and 6 months and is greater than the fellow eye, in which the improvement is slower but sustained over the 12 months. We saw the greatest post-treatment improvement in function predominately in the pre-treatment region of greatest dysfunction, irrespective of whether this was in the treated or untreated eye (Fig. 11). This improvement in the regions that we hypothesize are at greatest risk for developing advanced disease, provides the first piece of circumstantial evidence that the risk of progressing may have been reduced following 2RT laser treatment.

One outcome from this study has been the observation that patients with any atrophy, even when not apparent clinically but only seen on FAF, or in very small regions on the OCT, as missing RPE and photoreceptor layer, or even just a thin retina on OCT may not be able to be helped with this treatment. We also caution against treating large subfoveal drusenoid PEDs (>1000 μm) as two individuals lost vision over the follow-up period. We recommend that these cases should be excluded from future studies.

We occasionally observed a period after treatment, when there was rapid change in drusen load, where there was reduced visual function or acuity in some regions, but this was usually temporary. We believe this was due to a period of structural 'normalization' whereby the photoreceptors needed to re-align after a period of rapid change in the terrain associated with drusen resolution.

The aim of this pilot study of was to determine primarily, the safety of nanosecond laser in the setting of intermediate AMD and then to establish if there were change in fundus characteristics and retinal function that would give reason to pursue this laser as a potential therapeutic strategy in these high-risk cases of AMD. While there are many limitations to this study, the greatest being no comparative placebo group, we believe it achieved the aim of providing information to allow us to go forward with a larger, randomized clinical trial. This is now underway to further clarify the potential effects of the 2RT laser on AMD and the capability of macular perimetry to monitor disease progression.

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