Evaluation of blood flow on optic nerve head after pattern scan and conventional laser panretinal photocoagulation

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Abstract
To evaluate the changes in the blood flow on retina and the optic nerve head (ONH) after conventional laser treatment and to compare it to that after patterned scanning laser (PASCAL) treatment in patients with severe nonproliferative diabetic retinopathy (S-NPDR).

In this prospective, cross-sectional study, the blood flow on retina and the ONH was assessed by laser speckle flowgraphy using the mean blur rate (MBR) in 39 eyes with S-NPDR before, 1, 4, 8, 12 weeks after panretinal photocoagulation (PRP). Of 39 eyes, 17eyes with 17 patients treated by conventional laser and 22 eyes with 22 patients treated by PASCAL.

The mean age was 55.5 ± 11.5 years in the conventional laser group, 55.6 ± 11.8 years in the PASCAL group. The MBR-vessel, which can be dominantly expressed as retinal blood flow, was significantly reduced after PRP treated by conventional laser (P < .001), but did not change after PRP treated by PASCAL. The ratio of MBR-vessel to the baseline was significantly lower in the conventional laser group only at Week 1 (P = .045). The MBR-tissue, which can be dominantly expressed as the ONH blood flow, did not significantly change after PRP in the both group. The multiple stepwise regression analysis revealed that the laser burns was an independent factor significantly correlated with the ratio of MBR-vessel at Week 1 to the baseline (β = -0.550, P = .012).

The retinal blood flow was significantly reduced during the 12 weeks only after completion of PRP by conventional laser treatment. Our results indicate that short pulse on PRP treatment performed by the PASCAL would not significantly reduce the retinal blood flow.

Abbreviations: AU = arbitrary units, BCVA = best-corrected visual acuity, DBP = diastolic blood pressure, FFA = fluorescein fundus angiography, IOP = intraocular pressure, LSFG = laser speckle flowgraphy, MAP = mean arterial blood pressure, MBR = mean blur rate, NO = nitric oxide, OCT = optical coherence tomography, ONH = optic nerve head, OPP = ocular perfusion pressure, PASCAL = patterned scanning laser, PDR = proliferative diabetic retinopathy, PRP = panretinal photocoagulation, SBP = systolic blood pressure, SFCT = subfoveal choroidal thickness.

Keywords: laser speckle flowgraphy, mean blur rate, panretinal photocoagulation, patterned scanning laser, retinal blood flow

1. Introduction
Diabetic retinopathy is one of the leading causes of blindness in the industrialized world. In 1976, the diabetic retinopathy study showed that panretinal photocoagulation (PRP) for proliferative diabetic retinopathy (PDR) with high-risk characteristics decreased the risk of severe vision loss by over 50% out to 4 years of follow-up.[1] Thereafter, studies have demonstrated that PRP decreases the incidence of blindness in patients with PDR.[2,3] A 5-stage disease severity classification has been proposed for eyes with diabetic retinopathy; 3 stages of low risk, a fourth stage of severe non-PDR (S-NPDR), and a fifth stage of PDR.[4,5] The Early Treatment Diabetic Retinopathy Study reported that PRP treatment was associated with good long-term visual acuity in most patients with S-NPDR and PDR.[6]

There has been reported that a significant reduction in the retinal blood flow after PRP in eyes with PDR.[7-9] Grunwald et al demonstrated there were decreased venous diameter, flow velocity, and total blood flow following PRP.[7,8] Fujio et al demonstrated that after PRP treatment of half of the fundus, there were statistically significant regional decreases in the retinal blood flow (50%–78%) and decreased vessel diameters, ranging from 1% to 9%.[9]

Many techniques have been developed for measuring retinal flow including fluorescein angiography,[10] radioactive microspheres, hydrogen clearance, bidirectional laser Doppler velocimetry,[7,8] color Doppler ultrasonography,[11] and a pulsatile technique.[12] However, the change in retinal blood flow in detail after PRP has not been fully understood, presumably because of time-intensiveness and poor reproducibility in many techniques for measuring retinal blood flow.

Laser speckle flowgraphy (LSFG) is a noninvasive, real-time method that can measure the relative blood flow without the use of...
of contrast agents[13,14] and performs repeated measurements of
defined retinal regions, which makes it possible to quantitatively
estimate blood flow changes in retina and the optic nerve head
(ONH), in vivo. Blood flow can be quantified as the mean blur rate
(MBR) through the use of the LSFG-NAVI (Softcare Ltd,
Fukuoka, Japan).

Blumenkranz et al reported a patterned scanning laser
(PASCAL) treatment technique[15] that enabled a rapid applica-
tion of multiple laser spots in an array with short pulse durations
which decreases the width and the axial extent of the laser lesions
of the retinal pigment epithelium (RPE) and outer retinal layer.[16]
Accordingly, PASCAL treatment would have an advantage over
conventional laser therapy on the ocular blood flow. However,
there have been no publications showing the effect of PASCAL
treatment for DR on the retinal blood flow compared with
conventional laser treatment in a search of Pubmed. Therefore, a
comparison of the benefits of PASCAL and conventional laser
therapy on the retinal blood flow requires to be determined for
eyes with S-NPDR or PDR.

Thus, the aim of this study was to evaluate the changes in the
blood flow determined by LSFG on retina and the ONH after
conventional laser treatment and to compare it to that after
PASCAL treatment in patients with S-NPDR.

2. Methods

2.1. Ethics statement

This was a prospective, single-center, cross-sectional study, and
the procedures were approved by the Institutional Review Board
and the Ethics Committee of the Nagoya University Graduate
School of Medicine and registered with the University Hospital
Medical Network-clinical trials registry (UMIN000028453). The
procedures conformed to the tenets of the Declaration of
Helsinki. We explained the nature of the study to all patients
and obtained a signed informed consent.

2.2. Subjects

This study was conducted at the Nagoya University Hospital
between July 2016 and December 2017. Patients were randomly
assigned to conventional laser treatment or PASCAL treatment
by an independent investigator (YU) with an allocation ratio
of 1:1. All patients had a comprehensive ophthalmic examination
including measurements of axial length and intraocular pressure
(IOP), optical coherence tomography (OCT), slit-lamp exami-
nations, and fundus examinations before and 1, 4, 8, and 12 week
(s) after the PRP treatment. All patients were asked to abstain
from alcoholic and caffeinated beverages on the morning of the
examination. The pupil was dilated 30 minutes before the
examinations with tropicamide phenylephrine eye drops
(Mydrin-P, Santen Pharmaceutical, Osaka, Japan). The patients
rested for 15 to 20 minutes in a dark quiet room before the
examinations. We performed all examinations in the sitting position at approximately 12:00 hours to avoid diurnal varia-
tions.[17–19]

The IOP was measured with a handheld tonometer (Icare;
TiolatOy, Helsinki, Finland) and the axial lengths were measured
with a partial optical coherence interferometry (IOLMaster; Carl
Zeiss Meditec, La Jolla, CA). The diastolic blood pressure (DBP)
and systolic blood pressure (SBP) were measured with an
automatic sphygmomanometer (CH-48SC; Citizen, Tokyo,
Japan). The mean arterial blood pressure (MAP) and the ocular
perfusion pressure (OPP) were calculated as follows: MAP = DBP
+ 1/3 (SBP – DBP), and OPP = 2/3 MAP – IOP.

2.3. Exclusion criteria

The exclusion criteria included the presence of severe cataract,
vitreous hemorrhage, history of photocoagulation or intraretinal
surgery, retinochoroidal pathology, for example, choroidal
neovascularization, age-related macular degeneration, and
medical conditions that could influence the hemodynamics of
the eye other than diabetes, such as vascular diseases, hyperten-
sion, and arrhythmia.

2.4. PRP parameters

Fluorescein fundus angiography (FFA) demonstrated that all
patients had nonperfused retinal areas in 3 or more quadrants.
According to the recommendations of the Early Treatment
Diabetic Retinopathy Study group, PRP was performed in the
eyes with S-NPDR.[20] Topical anesthesia was used on all eyes
before using contact lens. The PRP was performed through a
widefield contact lens (Ocular Mainster PRP 165; Ocular
Instruments, Bellevue, WA).

The same surgeon (YM) performed all PRPs with a spot size of
200μm for both types of PRPs. A pulse duration of 20 ms was
used with the PASCAL treatment and 200 ms with the
conventional laser treatment.

In the conventional laser treatment, a 532-nm wavelength
argon laser device (NOVUS Varia; Lumenis, San Jose, CA) was
used. The power was set at 170 to 230 mW and the exposure was
maintained until a grayish-white lesion was seen to create an
effective retinal burn. The spots were placed at 1 spot distance
apart in the conventional group. The number of burns applied
during each session ranged from 300 to 400 spots and a total of
1200 to 1600 burns were applied to each eye. PRP was performed
in 4 sessions, the inferior, nasal, superior, and temporal
quadrants in that order at an interval of 1 week between
treatments.

A 5 x 5 square grid was used to treat 25 spots simultaneously
with PASCAL SlimLine (Topcon, Tokyo, Japan). We set the
power to 300 to 650 mW and performed PRP in 2 sessions at
precisely 1 week between the treatments.

2.5. Laser speckle flowgraphy

We used the LSFG-NAVI (Softcare Co, Ltd) instrument to
determine the ONH blood flow. The principles of LSFG have
been described in detail.[21–24] The MBR is a measure of the
relative blood flow velocity, and it is determined by examining the
pattern of the speckle contrast produced by the erythrocytes in
the ocular blood vessels.[13] The MBR images are acquired at a
rate of 30 frames/s over a 4-second period. The same site can be
measured by using the auto-tracking system. To evaluate the
circulation on the ONH, a circular marker was set surrounding
the eye other than diabetes, such as vascular diseases, hyperten-
sion, and arrhythmia.
of MBR-vessel and -tissue to the baseline was used to compare the effect of PRP between the conventional laser group and the PASCAL group.

2.6. Statistical analyses
Sample size was calculated with ONH MBR determined by LSFG in previous report, which showed 20\% reduction of ONH MBR after conventional PRP.\(^{[26]}\) At 90\% power and \(\alpha\) level of 0.05, a supposition that mean reduction of the ONH MBR after PRP is 20\% would need 17 subjects in each group, based on the reported standard deviation. Assuming a dropout rate of 20\%, 21 subjects in each group should be enrolled.

The values are presented as the mean ± standard deviation. Independent \(t\) test was used to compare normally distributed data. One way analysis of variance was used to evaluate changes in BCVA, CFT, MBR-vessel, -tissue, IOP, and OPP over time. Pearson correlation coefficient test was used to evaluate the association between the reduction ratio of MBR-vessel at Week 1 and independent variables, including laser burns, insulin/oral antidiabetic drugs, BCVA, eGFR, body mass index, creatinine, duration of diabetes, HbA1c, and age.

We performed all statistical analyses with SAS9.4 (SAS Inc, Cary). A \(P\) value <.05 was considered statistically significant.

3. Results

3.1. Patient demographics
A total of 46 patients with type II diabetes and S-NPDR without macula edema were screened and 43 patients were enrolled, 3 patients did not meet inclusion criteria. Of 43 patients, 21 patients were assigned to the conventional laser group and 22 patients to the PASCAL group. In the conventional laser group, 4 of 21 patients dropped out during PRP treatment. As a result, 17 eyes with 17 patients in the conventional laser group and 22 eyes with 22 patients in the PASCAL group were studied. No significant differences were observed in the systemic and ocular variables between the 2 groups (Table 1).

The mean number of the photocoagulation burns was 1524 ± 157 in the conventional laser group (Fig. 2) and 4959 ± 582 in the PASCAL group (Fig. 3). None of the patients developed any adverse events related to the PRP. The stage of the DR had not worsened as determined by the FFA findings at 12 weeks after the PRP.

3.2. Changes in ONH MBR
In the conventional laser group, the mean MBR-vessel was 39.5 ± 8.2 arbitrary units (AU) before PRP, 36.5 ± 10.1 AU (92\%) after the first session, 34.6 ± 9.0 AU (87\%) after the second session, 33.1 ± 7.4 AU (84\%) after the third session of the PRP, 29.0 ± 5.6 AU (76\%) at 1 week, 34.3 ± 11.5 AU (87\%) at 4 weeks, 34.3 ± 9.5 AU (87\%) at 8 weeks, and 36.9 ± 9.5 AU (92\%) at 12 weeks...

Table 1

| Characteristics                          | Conventional laser | PASCAL | \(P\)-value |
|------------------------------------------|--------------------|--------|-------------|
| n                                        | 17                 | 22     | .975        |
| Age, yr                                  | 55.5 ± 11.5        | 55.6 ± 11.8 | .975       |
| Males/females                            | 12/5               | 12/10  | .491        |
| BCVA (Log MAR)                           | 0.08 ± 0.10        | 0.09 ± 0.12 | .818       |
| Intraocular pressure, mm Hg              | 15.2 ± 2.7         | 15.0 ± 2.8 | .887       |
| Axial length, mm                         | 23.87 ± 0.73       | 23.91 ± 1.23 | .919       |
| Central foveal thickness, \(\mu\)m       | 227.4 ± 19.7       | 230.1 ± 26.4 | .747       |
| Subfoveal choroidal thickness, \(\mu\)m   | 283.2 ± 55.0       | 291.6 ± 41.1 | .922       |
| Systolic blood pressure, mm Hg           | 128.8 ± 32.1       | 131.6 ± 20.0 | .777       |
| Diastolic blood pressure, mm Hg          | 75.8 ± 18.7        | 78.3 ± 14.1 | .652       |
| Ocular perfusion pressure, mm Hg         | 47.1 ± 14.5        | 49.0 ± 12.8 | .681       |
| Heart rate, bpm                          | 81.8 ± 8.0         | 79.8 ± 14.3 | .627       |
| Duration of diabetes, yr                 | 12.6 ± 7.6         | 11.9 ± 8.1 | .802       |
| Insulin/oral antidiabetic drugs           | 10/7               | 12/10  | .953        |
| HbA1c (%)                                | 8.4 ± 1.9          | 9.3 ± 2.5 | .301       |
| Body mass index, kg/m²                   | 24.5 ± 5.0         | 24.2 ± 6.5 | .931       |
| Total cholesterol, mg/dL                 | 166.1 ± 43.1       | 175.5 ± 60.5 | .665       |
| Triglycerides (mg/dL)                    | 130.3 ± 48.1       | 134.1 ± 72.8 | .867       |
| Serum creatinine, mg/dL                  | 0.89 ± 0.29        | 0.75 ± 0.21 | .094       |
| eGFR, mL/min/1.73 m²                     | 69.3 ± 24.6        | 78.9 ± 27.3 | .273       |

(\(t\)-test)
after the completion of PRP (Table 2). The MBR-vessel was significantly decreased after PRP in the conventional group ($P < .001$) (Week 1, $P < .001$; Week 4, $P = .029$; Week 8, $P = .015$) (Fig. 4).

In the PASCAL group, the mean MBR-vessel was $39.1 \pm 7.4$ AU before PRP, $36.3 \pm 7.5$ AU (93%) after the first session, $34.9 \pm 8.9$ AU (87%) at 1 week, $34.9 \pm 8.2$ AU (87%) at 4 weeks, $37.4 \pm 6.6$ AU (96%) at 8 weeks, and $37.3 \pm 9.3$ AU (96%) at 12 weeks after PRP (Table 2). The MBR-vessel did not significantly change after PRP treatment in the PASCAL group.

The ratio of MBR-vessel to the baseline was significantly lower in the conventional laser group compared to the PASCAL group only at Week 1 ($P = .045$) (Fig. 4).

In the conventional laser group, the mean MBR-tissue was $10.3 \pm 2.8$ AU before PRP, $10.0 \pm 3.4$ AU (97%) after the first session, $9.5 \pm 3.1$ AU (93%) after the second session, $9.5 \pm 2.4$ AU (92%) after the third session of the PRP, $8.7 \pm 2.5$ AU (89%) at 1 week, $9.7 \pm 3.1$ AU (94%) at 4 weeks, $9.5 \pm 2.9$ AU (93%) at 8 weeks, and $10.1 \pm 3.3$ AU (98%) at 12 weeks following PRP (Table 2). The MBR-tissue did not significantly change after PRP treatment in the conventional laser group (Fig. 5).

In the PASCAL group, the mean MBR-tissue was $10.5 \pm 2.3$ AU before PRP, $10.3 \pm 2.3$ AU (98%) at the first session, $9.9 \pm 2.5$ AU (94%) at 1 week, $9.6 \pm 1.7$ AU (91%) at 4 weeks, $10.2 \pm 2.4$ AU (97%) at 8 weeks, and $10.4 \pm 2.7$ AU (99%) at 12 weeks following PRP (Table 2). The MBR-tissue did not significantly change after PRP treatment in the PASCAL group (Fig. 5).

The ratio of MBR-tissue to the baseline was not significantly different between the conventional laser group and the PASCAL group at any time (Fig. 1C).

3.3. Changes in other parameters

Although the BCVA, subfoveal choroidal thickness (SFCT), IOP, MAP, and OPP did not significantly change after PRP treatment in the both groups, the CFT significantly changed after PRP treatment in the conventional group ($P < .001$) and the PASCAL group ($P = .001$) (Table 2).

3.4. Correlations between ONH MBR and other parameters

Pearson correlation coefficient analyses showed that the ratio of MBR-vessel at Week 1 to the baseline was significantly correlated with the laser burns in the PASCAL group ($r = -0.506$, $P < .016$) (Table 3) (Fig. 5).
Figure 3. Fundus photographs and composite color maps of a 22-year-old woman with severe S-NPDR. Fundus photograph taken with Optos 200TX showing S-NPDR before (A) and after PRP treated by PASCAL (B). Representative composite color maps before, after first session, and 1, 4, 8, and 12 wk after PRP treatment on the ONH (C). There was no significant reduction in the MBR-vessel after the PRP. MBR = mean blur rate, ONH = optic nerve head, PASCAL = patterned scanning laser, PRP = panretinal photocoagulation, S-NPDR = severe nonproliferative diabetic retinopathy.

Table 2
Change in variable parameters with time.

| Parameter                  | Before PRP | Wk 1    | Wk 4    | Wk 8    | Wk 12   |
|----------------------------|------------|---------|---------|---------|---------|
| Conventional laser         |            |         |         |         |         |
| BCVA (Log MAR)             | 0.08±0.10  | 0.12±0.10| 0.11±0.11| 0.09±0.11| 0.09±0.10|
| CFT, µm                    | 227.4±19.7 | 240.5±30.0| 250.8±44.7| 252.8±36.3| 259.7±51.3|
| MBR-vessel (AU)            | 37.7±7.3   | 34.6±9.6 | 33.6±9.4 | 33.4±8.5 | 31.8±9.4 |
| MBR-tissue (AU)            | 9.6±2.4    | 9.6±3.3  | 9.6±3.0  | 9.6±3.1  | 9.4±2.9  |
| IOP, mm Hg                 | 15.1±2.7   | 13.4±2.4 | 13.9±2.7 | 13.4±1.9 | 13.7±2.0 |
| OPP, mm Hg                 | 47.1±14.5  | 48.5±13.5| 45.1±11.2| 48.4±7.7 | 48.4±8.5 |
| PASCAL                     |            |         |         |         |         |
| BCVA (Log MAR)             | 0.09±0.12  | 0.12±0.12| 0.11±0.13| 0.10±0.12| 0.09±0.13|
| CFT, µm                    | 230.0±29.0 | 239.6±40.0| 244.7±41.3| 246.4±32.5| 245.0±48.8|
| MBR-vessel (AU)            | 37.2±6.4   | 33.9±7.2 | 33.1±9.3 | 33.0±7.1 | 32.4±7.7 |
| MBR-tissue (AU)            | 9.9±2.3    | 9.9±1.5  | 9.7±1.8  | 9.4±2.9  | 9.4±1.8  |
| IOP, mm Hg                 | 15.0±2.8   | 13.6±2.8 | 13.7±3.1 | 14.3±2.8 | 14.5±2.9 |
| OPP, mm Hg                 | 49.0±13.1  | 47.2±9.1 | 47.9±9.2 | 46.2±9.2 | 46.2±10.8|

ANOVA = analysis of variance, BCVA = best-corrected visual acuity, CFT = central foveal thickness, IOP = intraocular pressure, LogMAR = logarithm of the minimum angle of resolution, MBR = mean blur rate, OPP = ocular perfusion pressure.
Figure 4. There was a significant reduction in the MBR-vessel after the PRP in the conventional laser group (A) and the ratio of MBR-vessel to the baseline in the conventional group was significantly lower than that in the PASCAL group at 1 wk after completion of PRP (B). There was not significantly reduction in the MBR-tissue in both groups (C) and no significant difference was observed in the ratio of the MBR-tissue throughout the post-PRP period (D). ∗∗∗P < .001, ∗∗P < .01, ∗P < .05. MBR = mean blur rate, PASCAL = patterned scanning laser, PRP = panretinal photocoagulation.

Figure 5. Scatterplot of the ratio of MBR-vessel at Wk 1 to the baseline versus the number of laser photocoagulation burns. The ratio of MBR-vessel at Wk 1 to the baseline was significantly correlated with the number of laser photocoagulation burns in the conventional laser group (\(r = -0.427, p = 0.086\)) (A) and the PASCAL group (\(r = -0.506, p = 0.016\)) (B). MBR = mean blur rate, PASCAL = patterned scanning laser.
The multiple stepwise regression analysis revealed that the laser burns was independent factors indicating the ratio of MBR-vessel at Week 1 to the baseline ($\beta = -0.550$, $P = .012$) (Table 4). The trend of the change in MBR-vessel and -tissue were not significantly correlated with that in the BCVA, SFCT, CFT, IOP, MAP, and OPP.

4. Discussion

Our results demonstrated a significant reduction of the MBR-vessel after the PRP only by the conventional laser treatment in eyes with S-NPDR. The reduction ratio of the MBR-vessel in the conventional laser group was significantly lower than that in the PASCAL group at 1 week following PRP. There were no significant correlations in the trend between the MBR-vessel or -tissue and other variables. The number of the laser burns was correlated with the ONH-MBR-vessel at 1 week after PRP.

Although many techniques have been developed for measuring retinal blood flow,[7,8,10–12] it is not easy to measure the retinal blood flow at the exact same area and evaluate the change in it after PRP treatment because of the shortcoming of time-intensiveness and poor reproducibility. Accordingly, we evaluated the retinal blood flow for the vascular areas of the ONH in eyes with S-NPDR and evaluated the change in it following PRP treatment. MBR-vessel on the ONH is separated from MBR using the “vessel extraction” function of the software, that is, which can be dominantly expressed as retinal blood flow.

There have been many reports that show a significant reduction in the retinal blood flow after PRP in eyes with PDR.[7–9,24–29] Grunwald et al demonstrated a reduction in venous diameter, flow velocity, and total blood flow following PRP.[7,8] Fujio et al demonstrated that there were statistically significant regional decreases in the retinal blood flow (50–78%) and decreased vessel diameters, ranging from 1% to 9% after PRP treatment of half of the fundus.[9] It has been demonstrated that the blood flow in severe NPDR with PRP on the ONH was significantly lower than that of severe NPDR without PRP using LSFG.[26]

There have been reported about the reasons for the reduced retinal blood flow after PRP as follows.[30–36] PRP improves the oxygenation of the ischemic inner retinal layers by destroying some of the metabolically highly active photoreceptor cells which would then lead to a greater flow of oxygen from the choriocapillaris to the inner layers of the retina.[30–32] Experimental studies show the improved oxygenation after retinal laser treatment,[33] and much higher retinal oxygen tension in laser treated areas than in untreated areas of the same retina.[34–36] When the changes in the oxygen flux reach the inner retina, the retinal arteries constrict, causing a reduction in the blood flow.[37–39]

In our study, the retinal blood flow was gradually decreased after every session of PRP in the both groups, and was significantly reduced after completion of PRP in the conventional group. In addition, the number of laser burns was significantly correlated with the reduction rate of retinal blood flow. These results indicate that PRP reduce the retinal blood flow and corroborate the previous reports[30–36] and the above mechanism.

The retinal blood flow was slightly recovered after the completion of PRP, which results that the retinal blood flow at Week 12 was not significantly different from the baseline in the conventional laser group. Recently, the similar tendency has been reported using LSFG.[40] Diddie et al found initial increase in retinal oxygen tension following retinal laser treatment but this effect was short-lived.[41] Photoreceptor outer segments damaged during photocoagulation in rabbits have been shown to recover by 4 weeks after laser irradiation.[42] The restoration of the photoreceptor layer in rats was more rapid, reaching its maximum extent by 3 weeks. The restoration can be observed in human eye using high-resolution OCT or OCT angiography.[43] In addition, Hiroshiiba et al reported that the leukocyte velocities in the retinal capillaries were significantly decreased immediately after photocoagulation and then gradually recovered to normal as time elapsed.[29] Those results suggest that the retinal blood flow is reduced immediately after laser photocoagulation as the above mechanism, but it can be recovered gradually with the restoration of the outer retinal layer, probably the increased retinal oxygen tension is reduced because of restoration of outer retina. On the other hand, there has been a study that evaluated 76 eyes with S-NPDR at 9 years following the PRP, and it was reported that the MBR determined on the ONH is significantly decreased at 70% of that of normal subjects.[26] Furthermore, the retinal vessels progressively constrict for several years following laser treatment.[44] Taken together, the possibility is not denied that the retinal blood flow is transiently recovered just following PRP treatment, but decreases gradually for a long period after PRP.

Table 3

| Variables                          | Conventional laser | PASCAL   |
|-----------------------------------|-------------------|----------|
|                                   | $r$               | $P$-value| $r$     | $P$-value |
| Laser burns                       | -0.427            | .086     | -0.506  | .016     |
| Insulin/oral antidiabetic drugs   | 0.004             | .989     | -0.345  | .116     |
| BCVA                              | 0.196             | .452     | -0.287  | .195     |
| HbA1c                             | 0.145             | .578     | 0.209   | .35      |
| Duration of diabetes              | 0.199             | .443     | -0.198  | .377     |
| Body mass index                   | 0.295             | .25      | 0.119   | .598     |
| Creatinine                        | -0.096            | .714     | -0.108  | .631     |
| eGFR                              | 0.006             | .981     | 0.094   | .678     |
| Age                               | 0.233             | .368     | -0.111  | .622     |
| Axial length                      | 0.01              | .968     | 0.046   | .838     |
| Foveal thickness                  | 0.092             | .724     | 0.08    | .725     |
| SFCT                              | 0.187             | .473     | 0.109   | .628     |

| Variables                          | $t$               | $P$-value |
|-----------------------------------|-------------------|----------|
|                                    |                   |          |
| BCVA                              | 0.138             | .498     |
| Age                               | 0.019             | .931     |

BOVA = best-corrected visual acuity, MBR = mean blur rate, SFCT = subfoveal choroidal thickness.

Table 4

| Variable                          | Independent       | $\beta$ | $P$-value |
|-----------------------------------|-------------------|---------|-----------|
| Reduction ratio of MBR-vessel at Wk 1 | Laser burns     | 0.550   | .012      |
|                                   | Insulin/oral antidiabetic drugs | -0.270 | .177      |
|                                   | BCVA             | -0.226  | .292      |
|                                   | eGFR             | -0.255  | .304      |
|                                   | Body mass index  | -0.082  | .408      |
|                                   | Creatinine       | 0.173   | .475      |
|                                   | Duration of diabetes | -0.141 | .489      |
|                                   | HbA1c            | 0.138   | .498      |
|                                   | Age              | 0.019   | .931      |

BCVA = best-corrected visual acuity, MBR = mean blur rate, PASCAL = patterned scanning laser.
The photocoagulation burns extended outside the laser lesioned area by at least 1 to 2 mm following the conventional laser treatment. \(^{145}\) PASCAL treatment has several advantages over conventional laser treatment including the short-pulse duration which decreases the axial extent and the width in the laser burns of the outer retinal layer and RPE. \(^{161}\) On the other hand, the effect of PASCAL treatment appears to be less than conventional laser treatment with the same number of laser spots. \(^{146}\) It has been reported that 6924 laser burns in an area of 8.36 mm\(^2\) is required in eyes with severe PDR and 3998 laser burns and an area of 456 mm\(^2\) is required in eyes with PDR to accomplish a complete regression of the disease using PASCAL treatment. \(^{147}\) Accordingly, a much larger number of laser burns with 4959 burns in the PASCAL treatment group was performed compared with 1524 laser burns in the conventional laser group.

Our results showed that the retinal blood flow was not significantly decreased during the 12 weeks following PRP by PASCAL. In addition, the reduction rate of retinal blood flow in the PASCAL group was significantly lower than that in the conventional group. These findings indicate that PASCAL therapy has a less effect on the retinal blood flow, even with enough number of photocoagulation burns until 12 weeks following PRP. Furthermore, the photocoagulation spot by PASCAL will not expand due to the short-pulse duration, which may then cause more differences in the retinal blood flow in the later period after PRP. On the other hand, Yamada et al found that retinal blood flow was significantly reduced during and after PRP treatments using PASCAL. \(^{140}\) The reason for this discrepancy between our and Yamada’s study was not determined. However, the response to PRP treatments varies among individuals, and difference of laser spot or power might have contributed to the lack of significant correlations.

The MBR-tissue was not significantly changed in the both groups. However, it has been reported that the MBR-tissue is significantly decreased at 79% of that of normal subjects at 9 years following the PRP. \(^{262}\) Also, studies have noted ONH pallor in diabetic eyes with PRP treatment, \(^{148,149}\) implying that blood flow is decreased in the tissue of the ONH. Taken together, it might be that blood flow in the tissue of the ONH does not reduce rapidly but continues to reduce for a long time after the PRP.

Limitations of this study included small sample size. In the conventional group, although the reduction ratio was not statistically significantly correlated with the laser burns probably because of small number, the correlation coefficient was −0.427 and those were though to be weak correlation. Second, the follow-up was short-term and we do not know about the change in retinal or ONH blood flow after 12 weeks after PRP. Third, the first post-laser examination was at 1 week in our study, and the change in retinal blood flow may have been occurred at an earlier period than 1 week. Further longitudinal studies using a larger number of subjects will be necessary for clarification.

In conclusion, the retinal blood flow was significantly reduced during the 12 weeks only after completion of PRP by conventional laser treatment. Our results indicate that short pulse on PRP treatment performed by the PASCAL would not significantly reduce the retinal blood flow.

**Author contributions**

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