Author's response to reviews

Title: Responsiveness of Eyes with Polypoidal Choroidal Vasculopathy with Choroidal Hyperpermeability to Intravitreal Ranibizumab

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Author's response to reviews: see over
Response to Reviewers’ comments:

To Reviewer 1:

This is an interesting paper that uses clinical interventions to determine the role of VEGF in PCV cases with and without choroidal hyper-permeability. Based on the response of these 2 different groups to anti-VEGF treatment, the authors make some interesting conclusions about the pathogenesis of PCV. I think that this study is worthy of publication, but I think it requires some modification. Mainly, the authors need to expand their discussion and conclusions to include the role of PDT in PCV. Also, the authors need to change their statistical analysis to account for the multiple comparisons that they did.

1. **The authors”conclusion is” Some type of PCV with HP may not be strongly associated with VEGF-related pathology.? I would agree with this, but I think that this conclusion can perhaps be broadened. These hyper-permeable cases that do not respond well to anti-VEGF monotherapy may benefit from combination therapy with PDT. I would suggest changing the conclusion to ?Cases of PCV that are associated with choroidal hyper-permeability may not be strongly associated with VEGF-related pathology, and may not respond favorably to anti-VEGF monotherapy?**

**Answer:** The conclusion was changed in the Abstract to, “Cases of PCV that are associated with choroidal hyper-permeability may not be strongly associated with VEGF-related pathology and may not respond favorably to anti-VEGF monotherapy.”

2. **In the Introduction, the authors state that 2 major therapies for PCV include anti-VEGF or PDT. However, they do not mention that there are many papers that have shown that combination therapy with BOTH anti-VEGF is widely used. Many people would consider combination therapy to be the standard-of-care for PCV. The authors should expand their Introduction to include the role of combination therapy, and cite the many papers that have described combination therapy for PCV.**

**Answer:** We agree with this suggestion. This section was revised with the additional of two references as follows.

**Page 3, line 10.**

“The major treatments for a subfoveal PCV are photodynamic therapy (PDT), intravitreal ranibizumab (IVR), an anti-vascular endothelial growth factor (VEGF) antibody, or a combination of PDT and anti-VEGF therapy [1,6-10]. The efficacy of IVR for PCV has been reported to be less than that for typical AMD [6,11,12].”

Additional references

8. Cho M, Barbazetto IA, Freund KB: Refractory neovascular age-related macular degeneration

3. **In the Introduction the authors state that PCV eyes with choroidal hyper-permeability may be more similar to eyes with CSC. It is important to note that PDT is the standard-of-care for CSC. I think the authors should expand on the role of PDT in CSC in the Discussion, and use this as an analogy to explain why combination therapy with PDT may be more appropriate in PCV eyes with choroidal hyper-permeability.**

**Answer:** The following sentences with additional references were added to the Discussion section.

Discussion section on page 8, last paragraph.

“Hyperpermeable choroidal vessels increase the tissue hydrostatic pressure, which overpowers the barrier functions of the RPE leading to serous retinal detachments [26]. There are various factors that cause the hyperpermeability of the choroid in CSC such as stasis, ischemia, inflammation, and other related factors [27]. At present, PDT is regarded to be the standard treatment for CSC [27]. Although the real mechanism of how PDT resolves the CSC has not been fully determined, it is assumed that PDT induces choroidal vascular remodeling with thinning of the choroid [28]. This effect was assumed to also occur in eyes with PCV by the frequent disappearance of the polypoidal lesions after PDT treatment [7].

On the other hand, anti-VEGF agents are not considered first-line treatments for CSC. Several small trials have yielded suggestive results, although their effectiveness has not been confirmed. Bae et al showed in a small randomized trials that half-fluence that PDT may be superior to anti-VEGF agent as a treatment for CSC [29]. Anti-VEGF agents are highly effective in reducing subretinal fluid, reducing extravasation from retinal vessels, and neovascularization, but they may not be sufficient to reduce the hyperpermeability of the choroid in eyes with CSC and PCV. The similarities between the PCV eyes with hyperpermeability and CSC eyes suggest that they may have the same pathogenesis and pathophysiology [6,10,11,27].

Lim et al reported that the responsiveness to bevacizumab as shown by the resolution of the subretinal fluid in eyes with CSC was better in eyes with than without hyperpermeability. Their results are just the opposite of our results [23]. However, Koizumi et al recently reported that the responsiveness to ranibizumab, expressed as resolution of retinal fluid in eyes with PCV, was worse in eyes with than without hyperpermeability which is consistent with our results [30]. Lim et al studied CSC and not PCV [23]. Although there may be similarities between the two diseases, they are not the same disease. Additionally, the definition of hyperpermeability in
CSC may not be the same as that of PCV. These factors could lead to the different response patterns after the treatment in the reports.”

Additional references.


4. The authors state that they examined records of all patients diagnosed between September 2010 and December 2011. Did they examine the records of all cases? Was this a consecutive series? It would be important to specify this in order to show that there was no bias in case selection.

Answer: We checked every medical record of patient with PCV. Therefore, this is a consecutive series. However among them, several cases were not included as was explained in the Results section; those with vitreous hemorrhage, hazy media, or poor quality of OCT images. Therefore, there was a potential bias in the case selection. This issue is explained in the Discussion section as follows.

“...was not a prospective randomized study but a retrospective study...”

5. The sentence “The patients were divided into a hyperpermeability group (HP group) and a normal choroidal permeability group (NP group) based on the ICGA findings with or without choroidal hyperfluorescence in the midphase of ICGA about 10 minutes after ICG injection.” could perhaps be clarified. Maybe change to “The patients were divided into a hyperpermeability group (HP group) and a normal choroidal permeability group (NP group) based on the ICGA finding of choroidal hyperfluorescence in the midphase of ICGA about 10 minutes after ICG injection.”

Answer: The sentence was changed as indicated on page 5, second paragraph.

6. ??and 100 scans were averaged.? Please clarify if this was A-scans or B-scans.
Answer: We used the OCT Star (enhanced depth imaging) protocol with 768 A Scan/B scan and 100 scans were averaged. This is described on Page 5, last line as follows.

“All of the images were obtained using the eye-tracking system. The OCT Star (enhanced depth imaging) protocol with 768 A scan/B scan and averaged 100 scans was used.”

7. The authors did multiple pair-wise comparisons, and did not adjust for multiple comparisons in their statistical analysis. Table 2 shows multiple pair-wise comparisons between the HP and NP groups. The authors need to account for the multiple comparisons in their statistical analysis in order to prove that their findings are not due to chance. Also, they need to adjust for the differences in the 2 groups that were shown in Table 1. A logistic regression model with multiple variables may be appropriate. Statistical consultation may be needed.

Answer: We consulted a professional biostatistician, Dr Takehiro Yamashita, MD, PhD, Kagoshima University, Japan, who studied at the Moorfields Eye Hospital, UK. He recommended that we present the results of the two analyses.

Because we analyzed the CFT 3 times, we had to use the Bonferroni’s correction. As a result, a statistically significant $P$ value should be $< 0.0167$. Even after Bonferroni’s correction, there was a significant reduction in the rate of CFT.

As the reviewer suggested, we also performed multivariate regression (stepwise) analysis on age, sex, permeability, CCT, laterality, baseline CMT, lens status, refraction, baseline logMAR BCVA. This allowed us to obtain the variables for the regression equations including permeability (HP or NP) and laterality. Our results showed that $R = 0.478$ with a $P$-value was 0.009 for permeability and was 0.042 for laterality. Thus, FA permeability of choroid is still significant for the reduction rate of the CFT.

This was described as follows.

Methods, page 6, last paragraph.

“Multivariate regression (stepwise) analysis was performed on age, sex, permeability, CCT, laterality, baseline CMT, lens status, refraction, and baseline logMAR BCVA.”

Page 9, last paragraph.

“Additionally, multivariate regression (stepwise) analysis was performed on age, sex, permeability, CCT, laterality, baseline CMT, lens status, refraction, and baseline logMAR BCVA. This allowed us to obtain the variables for the regression equations including permeability (HP or NP) and laterality. As a result, $R=0.478$ with a $P$-value of 0.009 for permeability and 0.042 for laterality. Thus, the FA permeability of choroid was still significant for the reduction of CFT.”
We found minor typographical error in Table 1. The $P$-value of contralateral eye involvement was 0.277 (3/13 in HP group and 7/14 in NP group). It was corrected in new Table 1.

8. The Discussion mentions similarities to CSC, and I think that at this point they should discuss how CSC responds to PDT and how this may be an argument for combination treatment is certain cases of PCV that are ?CSC-like?.

Answer: This comment and question was answered for question 3.

9. The authors discuss 2 different studies of CSC that showed conflicting results. They state “We cannot explain the differences but it may be related to the differences in the diseases examined.” I think a more detailed interpretation of the differences is needed.

Answer: It was rephrased as follows.

Page 9, last paragraph in the Discussion section.

“Lim et al reported that the responsiveness to bevacizumab as shown by the resolution of the subretinal fluid in eyes with CSC was better in eyes with than without hyperpermeability. Their results are just the opposite of our results [23]. However, Koizumi et al recently reported that the responsiveness to ranibizumab, expressed as resolution of retinal fluid in eyes with PCV, was worse in eyes with than without hyperpermeability which is consistent with our results [27]. Lim et al studied eyes with CSC and not with PCV. Although CSC and PCV have certain similarities, they are not the same disease. Additionally, the definition of hyperpermeability in CSC may not be the same as that of PCV. These factors might explain the different response patterns to the treatment between the reports.”

10. The authors would expand the Discussion to include the role of PDT in PCV, and describe how it may be appropriate as a combination with IVR in hyper-permeable cases that are resistant to IVR monotherapy.

Answer: It is described as follows on page 10, last paragraph.

“There have been recent reports on the results of combined PDT and anti-VEGF antibodies to treat PCV. Lai et al reported that IVR resulted in a stabilization of vision in patients with symptomatic PCV. However, combined IVR and PDT appeared to be more effective in causing a complete regression of the polypoidal lesions in ICGA compared with IVR monotherapy [33]. Others have shown the comparatively good results for maintaining the visual acuity [34,35]. Because there is no large scaled analysis of anti-VEGF monotherapy and combination therapy, the results are not conclusive. Considering our cases with PCV that are non-responsive to IVR, it might be meaningful to select specific cases requiring PDT rather than IVR (with or without PDT). The present finding would be helpful for that.”
11. The statement “The present method and interpretation, called interventional immunology, might be suitable for ocular diseases because the eye is a closed system and is less affected by the systemic circulation” should be modified, since the eye is not really a closed system. For example, intravitreal anti-VEGF therapy with bevacizumab has been shown to affect systemic VEGF levels for several weeks, suggesting that the system is not really “closed”.

Answer: The indicated phrases are corrected on page 11, second paragraph as follows.

“The present method and interpretation, called interventional immunology, might be suitable for ocular diseases because the eye is a relatively closed system and is less affected by the systemic circulation [18,19]. Although molecules injected into the vitreous can appear in systemic circulation, its effect is less than after an intravenous injection as is performed in cancer therapy.”

12. The Conclusion should be expanded to describe the potential role of combination therapy with PDT in these HP cases.

Answer: It was rephrased as on page 12, second paragraph as follows:

“This would suggest that the choroidal permeability in eyes with PCV is less dependent on VEGF-related pathology. For these eyes, combination therapy might be more suitable than IVR monotherapy.”

13. The authors? measured central choroidal thickness using EDI. I am assuming that this was done using the manual caliper function on the OCT software. Similar to the assessment of choroidal permeability on ICG, this is subjective and can vary between observers. The authors have stated that inter-observer agreement for assessment of choroidal permeability, but they have not done so for choroidal thickness. I would suggest that choroidal thickness using OCT-EDI be assessed by 2 or 3 different masked graders, and then the results averaged and the inter-observer agreement stated.

Answer: The OCT-EDI was done exactly as the reviewer indicated and as reported in our publication [21]. We describe the method as follows:

Methods, Page 5, last paragraph.
“Following each examination, the best image was projected on a computer screen and evaluated by 2 independent masked graders (NA and MS). Measurements of the choroidal thickness (rating) was done in a masked fashion by these 2 raters with no information on the eyes. The average of the results by two raters was used for the analyses of the choroidal thickness.”

Page 7, last line.

“The intra-class and inter-rater correlation coefficient using a two-way mixed effects model for measurements of absolute agreement were computed.”
Page 7, last paragraph.

“The inter-rater agreement of the choroidal thickness was very high with a coefficient of variance 0.932. The intraclass correlation coefficient had a 95% confidence interval of 0.887 to 0.964.”

14. There are some spelling and grammatical errors: ??this study were approval by the Institutional..?

Answer: The revised manuscript has been edited by native English speaking Professor Emeritus, Duco Hamasaki, of Bascom Palmer Eye Institute, University of Miami.

Reviewer 2

The authors describe 42 eyes with PCV and document the relationship of choroidal hyperpermeability with the response to anti-VEGF injections. This is an interesting topic as we learn more about the choroidal impact on retinal pathology with advanced imaging.

1) Elaboration of PCV as a distinct entity versus a variant of type 1 sub-RPE neovascular growth pattern would be pertinent. When reviewing the ICGs, did the polypoidal lesions in this series appear to be an outgrowth of pre-existing type 1 CNV or was it distinct?

Answer: This question of whether PCV is a variant or an outgrowth of type 1 CNV or abnormal dilated choroidal vessels has been discussed often. From our experience [2,5], there is a certain percentage of eyes with PCV that originate from abnormal choroidal vessels with apparent pulsatile motion. However, there are others that cannot be differentiated from type 1 CNV. These patterns were also noted in the present eyes. In this study, the diagnosis was made based upon the previous reports [1-6].

This issue is very important but may be beyond the scope of the present study. We appreciate this comment and will analyze this issue in the next study. This was described as follows.

Page 11, fourth paragraph.

“From the early days of PCV study, it was asked whether PCV originated from abnormal choroidal vessels or was a variant of type 1 CNV [4-6]. This issue might be related to the responsiveness to IVR. Further studies are needed.”


Answer: This reference was cited as follows.
“From the early days of PCV study, it was questioned whether PCV originated from abnormal choroidal vessels or was a variant of type 1 CNV [4-6]. This issue might be related to the responsiveness to IVR. Further studies with more advanced method such as simultaneous ICGA and eye-tracked SD-OCT would be helpful in obtaining an answer [39].”

Additional reference


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