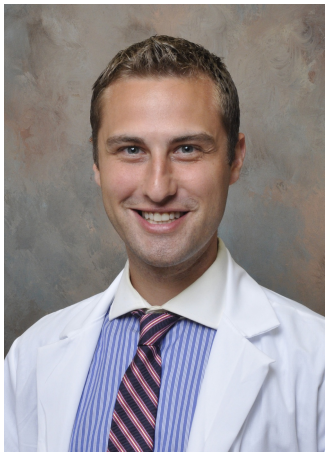
7/28/2020 11:15AM

Management of Proliferative Vitreoretinopathy with Intravitreal Methotrexate using a Treat-and-Extend Protocol



- Scott D Walter, MD, MSc

OBJECTIVE To report a novel dosing strategy using intravitreal methotrexate (MTX) as an adjuvant chemotherapy to prevent recurrent retinal detachment (RD) from proliferative vitreoretinopathy (PVR).

PURPOSE Previous investigators have demonstrated high rates of anatomic success using a series of 13 intravitreal MTX injections over 4 months. This study seeks to determine whether similar success can be achieved with fewer injections using a treat-and-extend protocol.

METHODS Retrospective consecutive case series of 22 eyes undergoing surgical repair of RD complicated by grade C or severe grade B PVR; including patients who received intraoperative and post-operative intravitreal MTX injections (400mcg/0.1mL); excluding patients with PVR secondary to proliferative diabetic retinopathy, sickle cell retinopathy, and ruptured globe injury. All patients underwent vitrectomy with membrane peeling and/or relaxing retinectomy of 180-360 degrees. The first dose of MTX was administered intraoperatively; subsequent doses were administered weekly until the retinectomy edge healed, then every 2-4 weeks until silicone oil (SO) was removed; a final dose was given at SO removal.

RESULTS The final reattachment rate was 100%. Two eyes (9%) required a single reoperation for recurrent RD with PVR. The median improvement in visual acuity was -0.9 logMAR. The cumulative intravitreal dose of MTX ranged from 2.0 - 5.2mg (median 2.8mg). The most common adverse event was vortex keratopathy which occured in 5 eyes (23%) during the weekly dosing phase, and resolved with extended dosing intervals.

CONCLUSION A high rate of anatomic success can be achieved using fewer MTX injections than in the previously described treatment protocol. The average patient in this series required 8 MTX injections: 2 intraoperatively and 6 in clinic over a period of approximately 2.6 months. Further study is needed to clarify the optimal MTX dosing frequency to prevent recurrent RD from PVR.

HUMAN RESEARCH Yes: Approved by institutional review board

7/28/2020 11:19AM

Clinicopathologic correlations of retinal membranes associated with intravitreal “stem cell” injections

- Rehan M. Hussain, MD
- Thomas A. Albini, MD
- Harry W. Flynn, MD
- Sander R Dubovy, MD
- Ajay E Kuriyan, MD, MS

OBJECTIVE What is the histologic composition of preretinal and subretinal membranes that form after non-FDA regulated intravitreal injection of autologous "stem cells"?

PURPOSE A growing number of for-profit, non-FDA regulated clinics offer unproven autologous “stem cell” injections (ASCIs) for ocular disease. There are several reports of profound vision loss following ocular injection of ASCIs, in association with tractional and rhegmatogenous retinal detachment (TRD/RRD), proliferative vitreoretinopathy (PVR), and epiretinal membrane (ERM) formation.

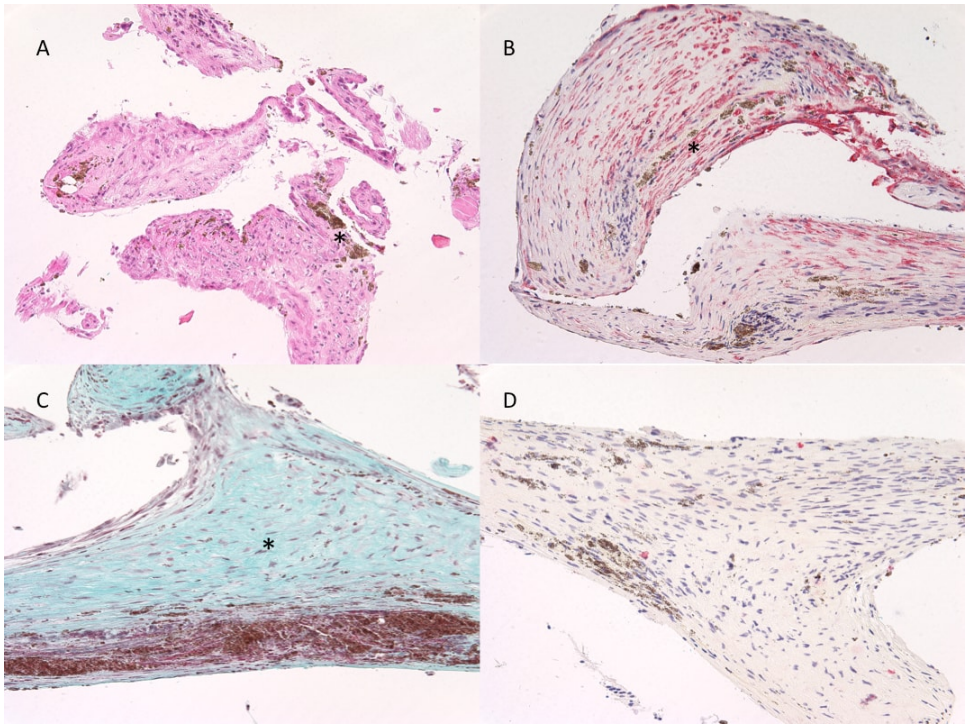
METHODS The histologic findings of membranes excised from 2 patients that developed RD after non-FDA-regulated ASCIs are reported. CASE 1:A 77-year-old woman with bilateral advanced wet AMD received intravenous and bilateral ASCI derived from her adipose tissue at an outside center. She developed TRD/RRD in the right eye 3 months after ASCI, for which she underwent two surgical repairs, with biopsy of a subretinal band. CASE 2:A 70-year old woman with autoimmune optic neuropathy received bilateral bone-marrow derived ASCI at an outside center. She developed extensive bilateral ERM 3 months later, followed by RRD with PVR in the left eye. The ERM was biopsied during RD repair.

RESULTS Case 1:Histologic evaluation of a subretinal band removed during surgery disclosed fibrovascular tissue that contained pigment-containing cells and spindle-shaped cells with fragments of glial tissue. Foci of dropout spaces were present (silicone oil). The membrane stained strongly positive with Smooth Muscle Actin (SMA) immunostain and positively for collagen deposition with Masson trichrome stain. CD34 immunostain (for hematopoietic cells) was negative. CD45 stain (common leukocyte antigen) was moderately

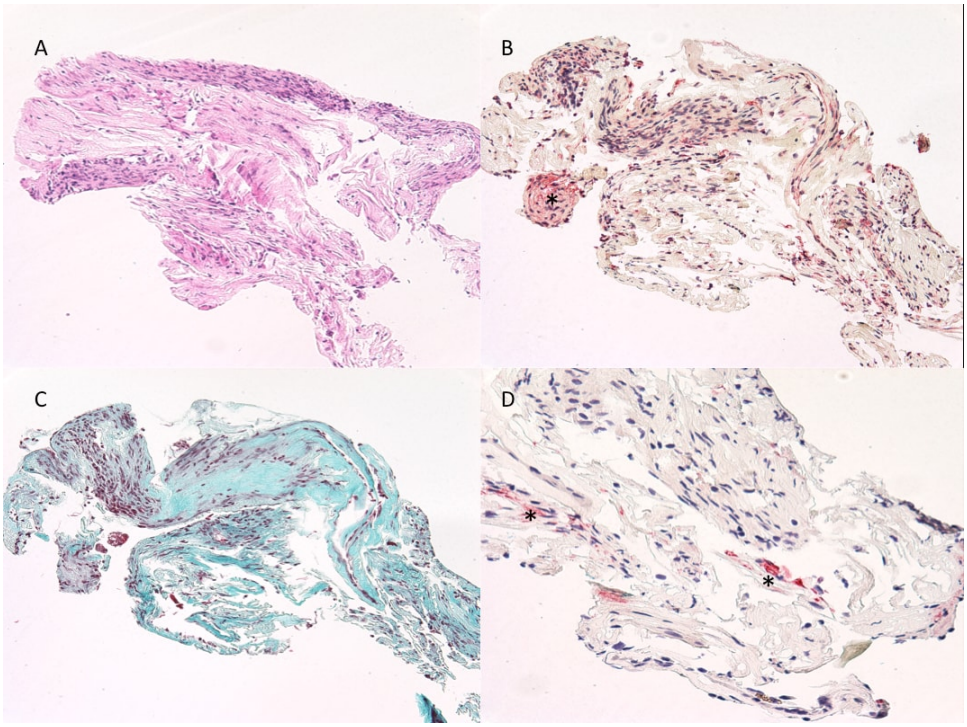
positive for inflammatory cells. Case 2:Histologic examination of the ERM disclosed periodic acid schiff-positive internal limiting membrane and a thick fibrocellular tissue present along one margin. The specimen stained positively for collagen deposition with Masson trichrome. SMA stained moderately positive, while CD34 (for hematopoietic cells) stained minimally positive.

CONCLUSION The specimens detailed in this report suggest that "stem cells," though present in one of two specimens, make up a minor component of these membranes, with the majority of the membranes composed of differentiated cells seen in typical PVR (which may include collagen, smooth muscle, glial and neuroendocrine cells, and RPE cells).

HUMAN RESEARCH Yes: Exempt from approval



(A) Histology of subretinal band demonstrates fibrovascular tissue that contained pigment-containing cells (asterisk) and spindle-shaped cells with fragments of glial tissue (presumed neural retina). Foci of dropout spaces were present, consistent with silicone oil. (B) The membrane stained strongly positive with Smooth Muscle Actin immunostain, colored red and marked with an asterisk. (C) The specimen was positive for collagen deposition with Masson trichrome, colored green and marked with an asterisk. (D) CD34 immunostain was negative.



(A) Histology of the epiretinal membrane from case 2 demonstrates a relatively thick fibrocellular tissue (hematoxylin-eosin, original magnification x200). (B) Smooth muscle actin stained moderately positive, colored in red and noted by an asterisk (original magnification x200). (C) The specimen stained positively for collagen deposition with Masson trichrome, colored green (original magnification x200). (D) CD34 immunostain was minimally positive, noted by an asterisk (original magnification x400).

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Outcomes of sub-silicone oil Triamcinolone acetonide application after 3600 relaxing retinectomy for complex retinal detachments - a comparative study

- Subhendu Kumar Boral, MBBS, MD(AIIMS), DNB(OPHTHALMOLOGY)
- Arnab Das, MD

OBJECTIVE To compare the anatomical and functional outcomes of with or without sub-silicone oil(SO) Triamcinolone acetonide crystal application after 360 degree retinectomy for complex retinal detachment.

PURPOSE To evaluate and compare the long term anatomical & functional outcomes of with or without application of sub-silicone oil additional Triamcinolone acetonide (TA) crystals after 360 degree relaxing retinectomy (RR) for complex retinal detachment (RD) with advanced proliferative vitreoretinopathy (PVR).

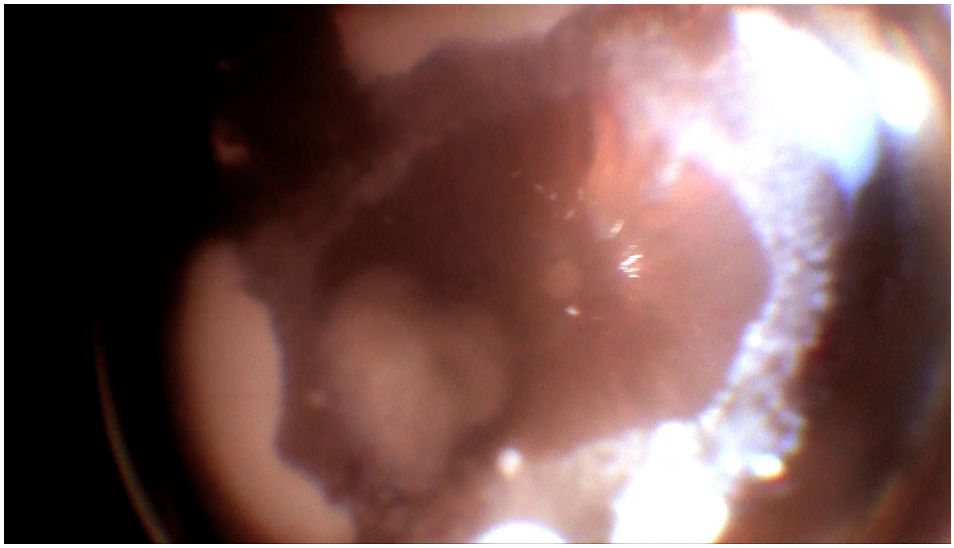
METHODS It was an institutional, retrospective, interventional case control study. TA assisted 23G or 25G vitrectomy was done in 24 complex retinal detachments with advanced PVR from various etiology where 360 degree RR had to be performed. Group A (n=13) included cases where additional TA crystal drops were applied over the site of RR after vitrectomy, PFCL, 360 degree RR & laser, before injecting silicone oil (SO 5000cst) tamponade or under it. In control arm group B (n=11), no additional TA crystals applied over RR sites under SO. Functional and anatomical outcomes were noted on follow up. Propensity to remove silicone oil was also measured as a secondary parameter.

RESULTS 16 were males and 8 females. Indications for 360 degree RR included complex rhegmatogenous retinal detachment with advanced PVR D2 or D3(n=12), severe anterior PVR (n=6) or 360 degree intrinsic retinal contraction (n=6). BCVA at presentation was invariably poor in both groups. Follow up was 12.46 ± 8.38 months in group A and 13.36 ± 9.23 months in group B. In group A, mean preoperative and post operative BCVA at final

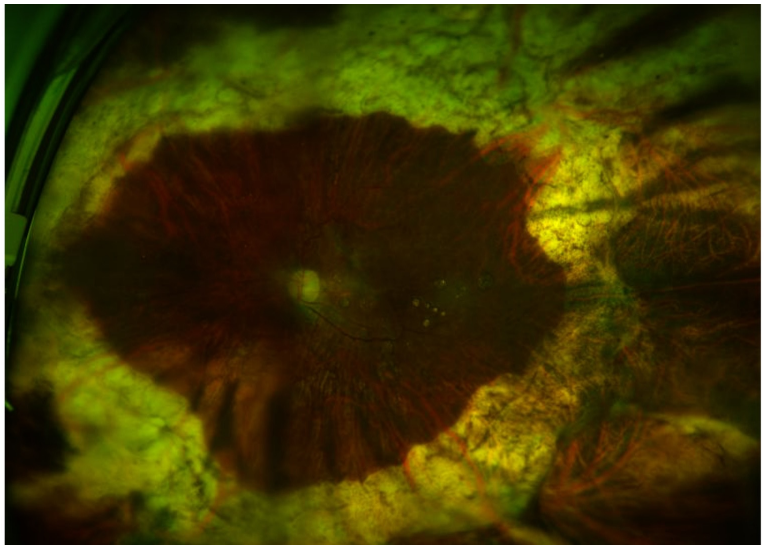
follow up were Log MAR 2.69 ± 0.41 and Log MAR 1.51 ± 0.90 ($p<0.05$) respectively. In group B, mean preoperative and post operative BCVA at final follow up were Log MAR 2.9 and Log MAR 2.37 ± 0.86 ($p<0.05$) respectively. Visual improvement in groupA was significantly better than group B (two sample t-test, $p<0.05$). Recurrence of RD developed in 1/13 cases in group A and 4/11 cases in group B (Fisher's Exact test, $p=0.002$, statistically significant). Silicone oil removal after 6 months of primary surgery was done significantly more in group A than group B (Fisher's Exact test, $p=0.0017$).

CONCLUSION Sub silicone oil TA crystals application over sites of RR after 360 degree relaxing retinectomy is a viable option in complex RD with extreme PVR. Trapped TA crystals in between silicone oil and bare RPE-Choroid exert localised anti-PVR effect. Visual improvement in groupA was significantly better with significantly less recurrence of RD. SO removal was significantly more recommended in group A

HUMAN RESEARCH Yes: Approved by institutional review board



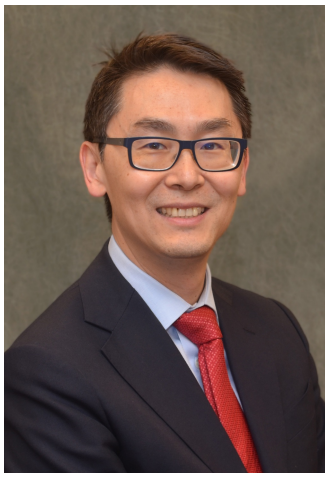
Intraoperative sub-silicone oil application of Triamcinolone acetonide crystal drops over the sites of 360 degree retinectomy in a case total old RD with 360 degree intrinsic retinal contraction.



On retina after silicone oil removal (after 6month of primary surgery) of the same patient

7/28/2020 11:33AM

Single Cell Genome-wide Expression Profiling of Patient-derived Membranes in Proliferative Vitreoretinopathy



- Leo A. Kim, MD, PhD
- Santiago Delgado-Tirado, MSc, MD
- Lucia Gonzalez-Buendia, MD, MSc
- Dhanesh Amarnani, MS
- Elizabeth Rossin, MD, PhD
- John B. Miller, MD
- Dean Eliott, MD
- Joseph Arboleda-Velasquez, MD, PhD

OBJECTIVE This study investigates the single cell gene expression of cells using single cell RNA-seq in patient-derived membranes in proliferative vitreoretinopathy.

PURPOSE To analyze single cell transcriptomes via single cell RNA-seq in order to characterize the cellular composition of proliferative vitreoretinopathy (PVR) membranes from human donors, and to investigate the role of the transcription factor RUNX1 in PVR.

METHODS Three PVR membranes were surgically obtained from three different patients with grade C PVR undergoing surgery. After tissue dissociation, PVR cells were sent for 3' v2 Single Cell Gene Expression analysis using the 10x Chromium Platform. Single cells, reagents, and a single Gel Bead containing barcoded oligonucleotides were encapsulated into nanoliter-sized GEMs (Gel Bead-in-Emulsion) using the GemCode platform. Lysis and barcoded reverse transcription of RNAs from single cells was performed. Full-Length barcoded cDNA was amplified by Polymerase Chain Reaction (PCR) to generated sufficient mass for library construction. Read counts were then analyzed using Partek Flow software.

RESULTS After computational filtering, 612 (87.55%), 758 (81.07%) and 1,026 (75.94%) cells remained, respectively within each sample. Within the 3 samples, 4 main clusters of cells were identified by differential gene expression. These clusters accounted for ≥90% of

the cells in each sample. RUNX1 expression was consistently expressed in the first two clusters across all three samples. Cell phenotyping distinguished one of the RUNX1-expressing cell clusters (Cluster 1), encompassing ~47.1% of all cells, as microglial cells due to their robust expression of hallmark genes of this cell lineage. This microglial population was found to be similar in size when compared to the other samples. Clusters 2 and 3 expressed gene sets characteristic of fibroblasts and mesenchymal cells, based on mean gene expression from published lists of genes characteristic of these cell types. These fibroblastic RUNX1-expressing cell clusters were correlated with mesenchymal gene expression.

CONCLUSION Single cell RNA-seq analysis confirmed the presence of a heterogeneous population of cells in PVR membranes and revealed the presence of immune and fibroblastic cell phenotypes in PVR. The majority of the cell clusters demonstrated robust RUNX1 expression, which correlated with a shift towards a mesenchymal phenotype, suggesting an essential role for RUNX1 in epithelial to mesenchymal transition.

HUMAN RESEARCH Yes: Approved by institutional review board

7/28/2020 11:37AM

Effect of etiology on the outcome in paediatric rhegmatogenous retinal detachment undergoing small gauge vitrectomy with silicon oil.

- Navneet Mehrotra, DNB
- Manish Nagpal, MD, FRCS (UK)
- Gayathri Mohan
- Sham Talati
- Akshar Soni, Dr

OBJECTIVE The effect of etiology on the outcome in paediatric rhegmatogenous retinal detachment undergoing small gauge vitrectomy with silicon oil.

PURPOSE To determine the functional and anatomical outcomes in paediatric rhegmatogenous retinal detachments (RRD) undergoing small gauge vitrectomy with silicon oil and assess the effect of etiology on the outcome.

METHODS Retrospective consecutive series of 49 eyes of children and young adults less than 18 years who underwent small gauge vitrectomy with silicon oil for RRD between 2016 and 2019. Minimum follow up of 6 months following silicon oil removal.

RESULTS Myopia was the commonest etiology (53.1%); Trauma (46.9%), Congenital or developmental (20.4%), previous surgery (16.3%). 71.4% of eyes had retinal reattachment at final follow up. Eyes with congenital anomalies had the best retinal reattachment rate at final follow up (80%) followed by myopia (73.1%). Previous surgery had least retinal reattachment rate (62.5%) and the worst functional outcome. Trauma cases showed best functional outcome.

CONCLUSION Myopia and trauma are the commonest etiologies in paediatric retinal detachment undergoing vitrectomy with silicon oil. Previous surgery is a prognostic factor for poor response. Congenital anomalies had the best retinal reattachment rate but not the best functional outcome.

HUMAN RESEARCH Yes: Approved by institutional review board