Drug treatment


Anti-VEGF Therapy for Neovascular AMD and Polypoidal Choroidal Vasculopathy.

Cheung GCM, Lai TYY, Gomi F, Ruamviboonsuk P, Koh A, Lee WK.

Abstract: Anti–vascular endothelial growth factor (anti-VEGF) therapy has revolutionized the treatment of neovascular age-related macular degeneration (AMD). This review will summarize the current evidence of anti-VEGF therapy in neovascular AMD, including subtypes of retinal angiomatous proliferation and polypoidal choroidal vasculopathy (PCV). Importantly, 2 large multicenter randomized clinical trials evaluating the safety and efficacy of anti-VEGF monotherapy and combination with photodynamic therapy (PDT) have recently reported initial first-year outcomes. In this review, we summarize the latest updates in the efficacy and safety of anti-VEGF monotherapy and combination with PDT in common lesion subtypes. Remaining gaps in current understanding are highlighted where further research is needed.

PMID: 28971633


Intravitreal Ziv-Aflibercept: Clinical Effects and Economic Impact.

Singh SR, Dogra A, Steward M, Das T, Chhablani J.

Abstract: During the past decade, drugs that inhibit the actions of vascular endothelial growth factor (VEGF) have become standard-of-care treatment for a variety of chorioretinal vascular conditions. The off-label, intravitreal use of ziv-aflibercept (Zaltrap) has provided clinicians with an additional cost-effective drug. The commercial preparation of ziv-aflibercept contains the same aflibercept (VEGF-trap) molecule as Eylea but has a much higher osmolarity (1000 mOsm/kg vs 300 mOsm/kg). Initial concerns regarding cytotoxicity and long-term safety of intravitreal ziv-aflibercept have been largely negated after a series of publications failed to identify adverse ocular and systemic side effects. Both treatment-naive and anti-VEGF–resistant cases of neovascular age-related macular degeneration (nAMD), diabetic macular edema (DME), retinal vein occlusion (RVO), and choroidal neovascular membrane (CNVM) may respond as well to ziv-aflibercept as to aflibercept. A higher dose of ziv-aflibercept (2 mg in 0.08 mL) does not cause any adverse effects during short-term follow-up period (1 month). Data from various sources suggest that ziv-aflibercept may be as cost effective as bevacizumab, thereby making it an attractive treatment option in low- and middle-income countries. However, problems with off-label use, compounding, and counterfeiting limit its availability in many countries. Data from prospective, randomized, multicenter clinical trials are still required to convince physicians and regulatory bodies of its clinical efficacy and potential as early therapy.

PMID: 28971631
Early Changes of Retinal Morphology in Therapy of Neovascular Age-Related Macular Degeneration with Three Commonly Used Anti-VEGF Agents.

Enders P, Sitnińska V, Altay L, Fauser S.

PURPOSE: To compare changes of retinal morphology in the first weeks following injection of anti-VEGF agents for neovascular age-related macular degeneration (nAMD).

PROCEDURES: In a prospective study 50 patients with active choroidal neovascularization secondary to nAMD were monitored weekly by spectral-domain optical coherence tomography for 3 weeks after treatment. Twenty-two patients received bevacizumab, 15 ranibizumab, and 13 aflibercept. Morphological parameters of retinal compartments were compared.

RESULTS: Mean central retinal thickness (391.22 ± 123.41 µm) was reduced by -26.15 µm (p < 0.001) after 1 week, by -12.54 µm (p < 0.001) after 2 weeks, and by -3.52 µm (p = 0.09) after 3 weeks. Mean intraretinal layer thickness changed only significantly between baseline and week 1 (p < 0.001). Mean subretinal thickness also decreased between weeks 1 and 2 (p = 0.01).

CONCLUSIONS: Early morphological changes occur primarily in the first 14 days after treatment. This information could be clinically helpful to evaluate early non-response.

PMID: 28950272

Dexamethasone Intravitreal Implant Therapy for Retinal Vein Occlusion Macular Oedema and Conversion to Ranibizumab in Clinical Practice.

Balal S, Than J, Tekriwal S, Lobo A.

PURPOSE: Evaluation of outcomes in retinal vein occlusions (RVOs) for: (1) multiple repeat dexamethasone (DEX) injections and (2) conversion from DEX to ranibizumab.

METHODS: We conducted a retrospective study evaluating outcomes of multiple DEX injections and those requiring conversion to ranibizumab at Moorfields Eye Hospital, Bedford, UK. All patients had undergone a complete ophthalmic work-up.

RESULTS: Patients (n = 129) had a mean follow-up of 19.9 months. The mean improvement in central retinal thickness was 312 µm after final DEX (p = <0.0001). Mean peak best corrected visual acuity (BCVA) after final DEX was an improvement of 16 ETDRS letters (p < 0.0001). Forty-nine patients were converted and received a mean of 9.37 ranibizumab injections with a mean improvement in BCVA of 15 ETDRS letters (p < 0.0001) compared with final DEX.

CONCLUSIONS: This study supports the use of ranibizumab in eyes previously treated with DEX and provides long-term efficacy and safety data for multiple DEX injection.

PMID: 28950262

Evaluating the Relationship Between Visual Acuity and Utilities in Patients With Diabetic Macular Edema Enrolled in Intravitreal Aflibercept Studies.


PURPOSE: The purpose of this study was to explore the relationship between visual acuity and utility...
METHODS: The relationship between visual acuity in the best-seeing eye (BSE) and worse-seeing eye (WSE) and utility was explored using ordinary least squares (OLS) and random-effects models adjusted for different covariates (age, age2, sex, body mass index, smoking status, glycated hemoglobin, diabetes severity, comorbidities, and geographic region). Utility was measured using the EuroQoL-five dimensions questionnaire (EQ-5D) and Visual Functioning Questionnaire-Utility Index (VFQ-UI). For each model, coefficients (R2) were reported, and WSE/BSE was expressed as the ratio of coefficients (OLS models). Models were independent of treatment effects, and outcomes from all time points (up to week 100) were included where available.

RESULTS: Data from 1320 patients with DME were analyzed. In all models, the association between visual acuity (BSE > WSE) was stronger with VFQ-UI than EQ-5D-derived utilities. The estimated relationship between VFQ-UI and visual acuity in the BSE and WSE was robust, even with an increasing number of covariates. WSE/BSE coefficient ratios were similar across VFQ-UI OLS models (32%) compared with EQ-5D models (41%-48%). Actual (unadjusted) versus predicted data plots also showed a better fit with VFQ-UI than EQ-5D-derived utilities.

CONCLUSIONS: These analyses show that VFQ-UI was more sensitive than EQ-5D-derived utilities for measuring the impact of visual acuity in the BSE and WSE. Visual acuity in the BSE was a major contributor to utility, but WSE is also important though to a lesser degree as shown by the coefficient ratios. These new data will be useful for health technology assessments in DME, where utilities data are lacking.

PMID: 28973328

Eur J Ophthalmol. 2017 Sep 14:0. [Epub ahead of print]

Aflibercept in the management of acute retinal necrosis syndrome-related macular edema.

Ortega-Evangelio L, Navarrete-Sanchis J, Williams BK, Tomás-Torrent JM.

PURPOSE: Acute retinal necrosis (ARN) is a panuveitis syndrome that may lead to severe complications such as cystoid macular edema (CME). There is no consensus about the best treatment. We report one case of CME secondary to ARN managed with intravitreal aflibercept.

CASE REPORT: A 41-year-old woman with a history of successfully treated varicella-zoster virus-associated ARN developed an epiretinal membrane (ERM) and underwent pars plana vitrectomy, ERM removal, inner limiting membrane peel, and lensectomy. After surgery, the retinal architecture improved and the visual acuity returned to 20/20. Six months later, she developed nontractional CME, which was treated monthly with triple-dose intravitreal aflibercept (2 mg). She gained 3 lines of vision and CME resolution was achieved.

DISCUSSION: Cystoid macular edema is a late complication of ARN that may affect vision. Some off-label therapies have been reported to be useful in CME secondary to ARN, including pegaptanib and interferon-α-2. Since interferon-α-2a is not currently available for ophthalmic use in Spain, aflibercept was the first choice. This soluble protein blocks the placental growth factor and all isoforms of vascular endothelial growth factor (VEGF); its half-life is prolonged and its affinity to VEGF-A is more than 100-fold greater than bevacizumab, pegaptanib, or ranibizumab. After each injection, macular thickness decreased consistently and visual acuity improved 3 lines after the treatment.

CONCLUSIONS: Intravitreal aflibercept is effective in the management of acute nontractional CME secondary to ARN.

PMID: 28967076
**Exp Eye Res. 2017 Sep 28. [Epub ahead of print]**

**Neutralization of placental growth factor as a novel treatment option in diabetic retinopathy.**

Van Bergen T, Hu TT, Etienne I, Reyns GE, Moons L, Feyen JHM.

Abstract: The current standard of care in clinical practice for diabetic retinopathy (DR), anti-vascular endothelial growth factor (VEGF) therapy, has shown a significant improvement in visual acuity. However, treatment response can be variable and might be associated with potential side effects. This study was designed to investigate inhibition of placental growth factor (PIGF) as a possible alternative therapy for DR. The effect of the anti-PIGF antibody (PL5D11D4) was preclinically evaluated in various animal models by investigating different DR hallmarks, including inflammation, neurodegeneration, vascular leakage and fibrosis. The in vivo efficacy was tested in diabetic streptozotocin (STZ) and Akimba models and in the laser induced choroidal neovascularization (CNV) mouse model. Intravitreal (IVT) administration of the anti-PIGF antibody was compared to anti-VEGFR-2 antibody (DC101), anti-VEGF antibody (B20), VEGF-Trap (aflibercept) and triamcinolone acetonide (TAAC). Vascular leakage was investigated in the mouse STZ model by fluorescein isothiocyanate labelled bovine serum albumin (FITC-BSA) perfusion and in the Akimba model by fluorescein angiography (FA). Repeated IVT administration of the anti-PIGF antibody reduced vascular leakage, which was comparable to a single administration of VEGFR-2 inhibition in the mouse STZ model. PL5D11D4 treatment did not alter retinal ganglion cell (RGC) density, as demonstrated by Bm3a staining, whereas DC101 significantly reduced RGC number with 20%. Immunohistological stainings were performed to investigate inflammation (CD45, F4/80) and fibrosis (collagen type 1a). In the CNV model, IVT injection(s) of PL5D11D4 dose-dependently reduced inflammation and fibrosis, as compared to PBS treatment. Equimolar single administration of the anti-PIGF antibody and aflibercept (21 nM) and TAAC decreased leukocyte and macrophage infiltration with 50%, whereas DC101 and B20 (21 nM) had no effect on the inflammatory response. Similar results were observed in the mouse STZ model on the number of microglia and macrophages in the retina. Repeated administration of PL5D11D4 (21 nM) and TAAC similarly reduced fibrosis, while no effect was observed after equimolar DC101, B20 nor aflibercept administration (21 nM). In summary, the anti-PIGF antibody showed comparable efficacy as well-characterized VEGF-inhibitor on the process of vascular leakage, but differentiates itself by also reducing inflammation and fibrosis, without triggering a neurodegenerative response.

PMID: 28965804

**J Ocul Pharmacol Ther. 2017 Sep 27. [Epub ahead of print]**

**Comparison of Strategies of Treatment with Ranibizumab in Newly-Diagnosed Cases of Neovascular Age-Related Macular Degeneration.**

Garweg JG, Niderprim SA, Russ HM, Pfister IB.

PURPOSE: In several case-studies improved outcomes have been reported after switching from a pro-re-nata (PRN)- to a treat-and-extend (T&E)-based therapeutic approach in cases of neovascular age-related macular degeneration (nAMD). We therefore wished to compare the effects of instigating 2 different protocols in newly-diagnosed nAMD undergoing treatment with Ranibizumab.

METHODS: The outcomes of a PRN- and a T&E-based regime were retrospectively compared in treatment-naïve eyes under therapy with Ranibizumab for minimally 12 months in a routine clinical setting. The primary outcome measures included the proportion of the eyes with intraretinal fluid in OCT and visual stability after the initial drug-loading phase.

RESULTS: The comparative case-series included 107 eyes (PRN: 68; T&E: 39). During the 2-year follow-up period, a similar number of clinical examinations were performed in the 2 groups (PRN: 14.0 ± 6.2; T&E 13.4 ± 4.4; P = 0.97), whereas the number of injections that were administered differed for the first (PRN: 5.5 ± 2.0 vs. T&E 6.8 ± 2.4; P = 0.008) and the second year (PRN: 1.9 ± 2.0 vs. T&E 3.8 ± 2.3; P = 0.002). The proportion of eyes with intraretinal fluid after the initial drug-loading phase remained stable (PRN: from 33.8% to 36.4%; T&E: from 25.6% to 29.0%); so, too, did the central retinal thickness and the visual acuity.
CONCLUSION: Despite a limited sample size, this retrospective analysis revealed the anatomical and the functional improvements during the 2-year follow-up period to be not roughly different for the 2 strategies. However, when the PRN-approach is instigated, the risk of under-treatment due to lapses in visits or to over-extensions in the intervals between treatments may be underestimated.

PMID: 28953427

Ophthalmology. 2017 Sep 27. [Epub ahead of print]

Clinicopathologic Correlation of Anti-Vascular Endothelial Growth Factor-Treated Type 3 Neovascularization in Age-Related Macular Degeneration.


PURPOSE: To correlate histologic results with previously recorded multimodal imaging results from a patient with type 3 neovascularization secondary to age-related macular degeneration (AMD).

DESIGN: Case study, clinical imaging, laboratory imaging, and eye-tracked clinicopathologic correlation.

PARTICIPANT: An 86-year-old white woman with type 3 neovascularization secondary to AMD treated with 6 intravitreal injections of bevacizumab.

METHODS: Multimodal retinal imaging at each clinic visit was correlated with ex vivo and high-resolution histologic images of the preserved donor eye. Clinical imaging included serial near-infrared reflectance and eye-tracked spectral-domain optical coherence tomography (OCT). Eye tracking, applied to the donor eye, enabled identification of histologic features corresponding to clinical OCT signatures.

MAIN OUTCOME MEASURES: Histologic correlates for clinical OCT signatures were sought, including reflectivity of the vascular complex, intraretinal hyperreflective foci and intraretinal cellularity, analysis of the topography of pathologic features, and evaluation of the sub-retinal pigment epithelium (RPE) plus basal lamina (BL) space.

RESULTS: Clinical imaging showed a deep neovascular lesion in close relationship with a mixed serous and drusenoid pigment epithelium detachment (PED), characteristic of type 3 neovascularization. Antiangiogenic therapy achieved a complete resolution of exudation. The PED progressively flattened with each treatment, leaving a persistent triangular hyperreflectivity in the outer retina. This persistent deep lesion histologically correlated with a vascular complex implanted into sub-RPE basal laminar deposit. No connection between the choriocapillaris and the sub-RPE plus BL space was observed. Both RPE-derived and lipid-filled cells were correlated with clinical intraretinal hyperreflective foci. The sub-RPE plus BL space contained macrophages, lymphocytes, Müller cell processes, and subducted RPE.

CONCLUSIONS: Clinicopathologic correlation of type 3 neovascularization showed vascular elements of retinal origin accompanied by collagenous material and Müller cell processes implanting into thick sub-RPE basal laminar deposit, which may simulate the appearance of chorioretinal anastomosis. Surrounding RPE-derived and lipid-filled cells thought to be microglia correlated with clinical intraretinal hyperreflective foci.

PMID: 28964579


[Summarize drug dosage forms in treatment of age-related macular degeneration disease]. [Article in Chinese]

Du MB, Liu SZ, Xu K, Liang LN, He AP, Yao Y, Liu YM.

Abstract: In this review, the authors summarized the drugs in treatment of the age-related macular
degeneration (AMD or ARMD), including the pathogenesis of the age-related macular degeneration at home and abroad, dosage forms used in the treatment, and the drugs research and development directions in the future. AMD disease is the third largest blinding diseases all over the world, with an incidence of 6.62%. The dosage form of the traditional medicine is mostly oral formulations, playing a role in body, while the newly dosage form is topical drug delivery formulation. Traditional Chinese medicine (TCM) has certain advantages in the treatment of AMD disease and the development of topical drug delivery preparations with newly preparation technologies would have a very bright prospect in the future.

PMID: 28959828


Alternative ways to optimize treatment for retinal vein occlusion with peripheral capillary non-perfusion: a pilot study.

Tultseva SN, Astakhov YS, Novikov SA, Nechiporenko PA, Lisochkina AB, Ovnanyan AY, Astakhov SY.

PURPOSE: We compared the efficacy and safety of ranibizumab versus ranibizumab plus scatter laser photocoagulation (SLP) in patients with chronic post-central retinal vein occlusion (CRVO) macular edema (ME).

METHODS: This prospective non-randomized pilot study included 250 patients with peripheral retinal ischemia and CRVO-related ME. The mean follow-up period was 24.5 ± 6.5 months. The clinical assessments conducted included best corrected visual acuity, optical coherence tomography, and multi-field fluorescein angiography with measurement of the ischemic area. The study population comprised two comparable patient groups with peripheral retinal ischemia that received different treatments for post-CRVO ME: ranibizumab with peripheral SLP of capillary non-perfusion areas (Group 1); and Lucentis® monotherapy (Group 2). Data analyses were performed using Statistica 7 software suite and included the estimation of \( \bar{x} \pm \delta \) values and their dispersion and covariation coefficients at different stages of the study.

RESULTS: Clinically significant retinal ischemia was detected in 175 (70%) patients, occupying an average of 435.12 ± 225.13 mm², i.e., 167.15 ± 45.16 optic disc areas. Peripheral ischemia was found in 125 patients, representing 50% of all patients with CRVO and 71.4% of all patients with ischemic CRVO. The mean number of ranibizumab injections in patients who underwent SLP was 3.5 ± 1.6. Patients treated with ranibizumab monotherapy for 24 months received 10.6 ± 2.5 injections. Functional and anatomic results were comparable in the two groups.

CONCLUSIONS: The combination of ranibizumab injections and peripheral SLP in capillary non-perfusion areas can significantly decrease the number of injections and reduce neovascular complications.

PMID: 28954021

Other treatment & diagnosis


Alterations in the Choriocapillaris in Intermediate Age-Related Macular Degeneration.

Borrelli E, Uji A, Sarraf D, Sadda SR.

PURPOSE: The purpose of this study was to compare the choriocapillaris plexus in eyes with intermediate AMD (iAMD), with or without neovascular AMD in the fellow eye, using optical coherence tomography angiography (OCTA).

METHODS: We collected data from 42 eyes with iAMD from 42 patients who had obtained OCTA. This cohort was divided into two subgroups according to the status of the fellow eye, yielding a group of 20
cases with bilateral intermediate AMD (bilateral iAMD group) and 22 cases with neovascular AMD in the fellow eye (unilateral iAMD group). An additional control group of 20 eyes from 20 healthy subjects was included for comparison. Main outcome measures were: (1) the percent of nondetectable perfused choriocapillaris area and (2) the average choriocapillaris signal void size.

RESULTS: No differences in the percent of nondetectable perfused choriocapillaris area were found among the three groups (2.3 ± 1.4% in the unilateral iAMD group, 1.5 ± 0.9% in the bilateral iAMD group, and 1.7 ± 1.4% in the control group, respectively). The average choriocapillaris signal void size, however, was significantly increased in unilateral iAMD eyes (293.7 ± 71.2 μm2) compared to both bilateral iAMD (241.5 ± 51.6 μm2, P = 0.031) and control (212.7 ± 48.6 μm2, P = 0.001) eyes.

CONCLUSIONS: Intermediate AMD eyes of patients with neovascular AMD in the fellow eye have an increased average choriocapillaris signal void size compared to eyes without neovascular AMD in the fellow eye. If replicated in future studies, choriocapillaris signal void size may prove to be a useful parameter for evaluating eyes with AMD.

PMID: 28973325


Automated Grading of Age-Related Macular Degeneration From Color Fundus Images Using Deep Convolutional Neural Networks.

Burlina PM, Joshi N, Pekala M, Pacheco KD, Freund DE, Bressler NM.

IMPORTANCE: Age-related macular degeneration (AMD) affects millions of people throughout the world. The intermediate stage may go undetected, as it typically is asymptomatic. However, the preferred practice patterns for AMD recommend identifying individuals with this stage of the disease to educate how to monitor for the early detection of the choroidal neovascular stage before substantial vision loss has occurred and to consider dietary supplements that might reduce the risk of the disease progressing from the intermediate to the advanced stage. Identification, though, can be time-intensive and requires expertly trained individuals.

OBJECTIVE: To develop methods for automatically detecting AMD from fundus images using a novel application of deep learning methods to the automated assessment of these images and to leverage artificial intelligence advances.

DESIGN, SETTING, AND PARTICIPANTS: Deep convolutional neural networks that are explicitly trained for performing automated AMD grading were compared with an alternate deep learning method that used transfer learning and universal features and with a trained clinical grader. Age-related macular degeneration automated detection was applied to a 2-class classification problem in which the task was to distinguish the disease-free/early stages from the referable intermediate/advanced stages. Using several experiments that entailed different data partitioning, the performance of the machine algorithms and human graders in evaluating more than 130,000 images that were deidentified with respect to age, sex, and race/ethnicity from 4613 patients against a gold standard included in the National Institutes of Health Age-Related Eye Disease Study data set was evaluated.

MAIN OUTCOMES AND MEASURES: Accuracy, receiver operating characteristics and area under the curve, and κ score.

RESULTS: The deep convolutional neural network method yielded accuracy that ranged between 88.4% (SD, 0.5%) and 91.6% (SD, 0.1%), the area under the receiver operating characteristic curve was between 0.94 and 0.96, and κ (SD) between 0.764 (0.010) and 0.829 (0.003), which indicated a substantial agreement with the gold standard Age-Related Eye Disease Study data set.

CONCLUSIONS AND RELEVANCE: Applying a deep learning-based automated assessment of AMD from fundus images can produce results that are similar to human performance levels. This study demonstrates
that automated algorithms could play a role that is independent of expert human graders in the current management of AMD and could address the costs of screening or monitoring, access to health care, and the assessment of novel treatments that address the development or progression of AMD.

PMID: 28973096


Fluorescence Lifetimes of Drusen in Age-Related Macular Degeneration.

Dysli C, Fink R, Wolf S, Zinkernagel MS.

PURPOSE: The purpose of this study was to characterize fundus autofluorescence lifetimes of retinal drusen in patients with AMD.

METHODS: Patients with AMD and retinal drusen and healthy controls of similar age were examined. A fluorescence lifetime imaging ophthalmoscope was used. Retinal autofluorescence was excited using a 473-nm pulsed laser, and fundus autofluorescence lifetimes of the central retina (30°) were measured in two distinct spectral channels (short: 498 to 560 nm [SSC]; long: 560 to 720 nm [LSC]). Mean retinal autofluorescence lifetimes, corresponding fundus autofluorescence intensity images, spectral domain optical coherence tomography, color fundus images, and clinical data were investigated. Patients were analyzed in two distinct groups (soft drusen and reticular pseudodrusen) and compared with control subjects.

RESULTS: Sixty-four eyes of 64 patients with AMD and retinal drusen (age: mean ± SD, 78 ± 8.5 years; range, 59 to 94 years) were investigated and compared with a control group of 20 age-matched healthy subjects. Mean retinal autofluorescence lifetimes in patients with AMD was significantly prolonged compared with the healthy control eyes (mean ± SEM: SSC, 486 ± 18 vs. 332 ± 11 ps, P < 0.0001; LSC: 493 ± 9 vs. 382 ± 17 ps, P < 0.0001). Areas of drusen featured a wide range of fluorescence lifetime values. Long lifetimes were identified in areas of atrophy and in areas of intraretinal hyperefficient deposits. Short lifetimes corresponded to deposits within the photoreceptor outer segment band.

CONCLUSIONS: Mean retinal autofluorescence lifetimes in AMD patients are significantly prolonged. Intraretinal deposits cause prolonged lifetimes, whereas deposits in the area of the outer photoreceptor segments lead to short fluorescence lifetimes.

PMID: 28973332

Ophthalmology. 2017 Sep 27. [Epub ahead of print]

Natural History of Subclinical Neovascularization in Nonexudative Age-Related Macular Degeneration Using Swept-Source OCT Angiography.


PURPOSE: Swept-source (SS) OCT angiography (OCTA) was used to determine the prevalence, incidence, and natural history of subclinical macular neovascularization (MNV) in eyes with nonexudative age-related macular degeneration (AMD).

DESIGN: Prospective, observational, consecutive case series.

PARTICIPANTS: Patients with intermediate AMD (iAMD) or geographic atrophy (GA) secondary to nonexudative AMD in 1 eye and exudative AMD in the fellow eye.

METHODS: All patients were imaged using both the 3×3 mm and 6×6 mm SS OCTA fields of view (PLEX Elite 9000; Carl Zeiss Meditec, Inc, Dublin, CA). The en face slab used to detect the MNV extended from
the outer retina to the choriocapillaris, and projection artifacts were removed using a proprietary algorithm.

MAIN OUTCOME MEASURES: Prevalence of subclinical MNV and time to exudation with Kaplan-Meier cumulative estimates of exudation at 1 year.

RESULTS: From August 2014 through March 2017, 160 patients underwent SS OCTA (110 eyes with iAMD and 50 eyes with GA). Swept-source OCTA identified subclinical MNV at the time of first imaging in 23 of 160 eyes, for a prevalence of 14.4%. Six eyes demonstrated subclinical MNV during the follow-up. Of 134 eyes with follow-up visits, a total of 13 eyes demonstrated exudation, and of these 13 eyes, 10 eyes were found to have pre-existing subclinical MNV. By 12 months, the Kaplan-Meier cumulative incidence of exudation for all 134 eyes was 6.8%. For eyes with subclinical MNV at the time of first SS OCTA imaging, the incidence was 21.1%, and for eyes without subclinical MNV, the incidence was 3.6%. There was no difference in the cumulative incidence of exudation from pre-existing MNV in eyes with iAMD or GA (P = 0.847, log-rank test). After the detection of subclinical MNV, the risk of exudation was 15.2 times (95% confidence interval, 4.2-55.4) greater compared with eyes without subclinical MNV.

CONCLUSIONS: By 12 months, the risk of exudation was greater for eyes with documented subclinical MNV compared with eyes without detectable MNV. For eyes with subclinical MNV, recommendations include more frequent follow-up and home monitoring. Intravitreal therapy is not recommended until prospective studies are performed.

PMID: 28964581


Green emission fluorophores in eyes with atrophic age-related macular degeneration: a colour fundus autofluorescence pilot study.


BACKGROUND/AIMS: To investigate the presence of short-wave fluorophores within regions of age-related macular degeneration (AMD)-associated macular atrophy (MA) area.

METHODS: This is a prospective, observational, cross-sectional case series. 25 eyes (18 patients) with late AMD and clinically identified MA were enrolled. Eyes were imaged using a confocal light-emitting diode blue-light fundus autofluorescence (FAF) device (EIDON, CenterVue, Padua, Italy) with 450 nm excitation wavelength and the capability for ‘colour’ FAF imaging, including both the individual red and green components of the emission spectrum. To produce images with a high contrast for isolating the green component, the red component was subtracted from the total FAF image. The main outcome measure was the presence of green emission fluorescence component (GEFC) within the MA area. Volume spectral domain optical coherence tomography (SD-OCT) scans were obtained through the macula and the OCT was correlated with the MA lesions identified on the FAF images, including regions of increased GEFC.

RESULTS: Of the investigated eyes, 11 out of 25 (44.0 %) showed the absence of GEFC in the MA area, whereas 14 eyes (56.0%) were characterised by GEFC within the MA area. The presence and distribution of GEFC in the MA area correlated with the presence of hyper-reflective material over Bruch’s membrane on the corresponding SD-OCT scans.

CONCLUSION: Short-wave fluorophores, which contribute to the GEFC, are present in the MA area and appear to correspond to residual debris or drusenoid material. Short-wavelength fluorophores revealed by colour FAF imaging may warrant further study.

PMID: 28972030
Retinal imaging in human autopsy eyes using a custom optical coherence tomography periscope.

McNabb RP, Tian J, Farsiu S, Izatt JA, Lad EM, Kuo AN.

Abstract: Age-related macular degeneration (AMD) is a major cause of vision loss in the elderly. To better study the pathobiology of AMD, postmortem eyes offer an excellent opportunity to correlate optical coherence tomography (OCT) imaging characteristics with histopathology. However, postmortem eyes from autopsy present challenges to standard OCT imaging including opaque anterior segment structures and standard of care autopsy processing resulting in oblique views to the macula. To overcome these challenges, we report a custom periscope attached by a standard mount to an OCT sample arm and demonstrate high quality macular OCT acquisitions in autopsy-processed eyes.

PMID: 28966854 PMCID: PMC5611930

A method for volumetric retinal tissue oxygen tension imaging.

Felder AE, Wanek J, Teng PY, Blair NP, Shahidi M.

PURPOSE: Inadequate retinal oxygenation occurs in many vision-threatening retinal diseases, including diabetic retinopathy, retinal vascular occlusions, and age-related macular degeneration. Therefore, techniques that assess retinal oxygenation are necessary to understand retinal physiology in health and disease. The purpose of the current study is to report a method for the three-dimensional (3D) imaging of retinal tissue oxygen tension (tPO2) in rats.

METHODS: Imaging was performed in Long Evans pigmented rats under systemic normoxia (N = 6) or hypoxia (N = 3). A vertical laser line was horizontally scanned on the retina and a series of optical section phase-delayed phosphorescence images were acquired. From these images, phosphorescence volumes at each phase delay were constructed and a 3D retinal tPO2 volume was generated. Retinal tPO2 volumes were quantitatively analyzed by generating retinal depth profiles of mean tPO2 (MtPO2) and the spatial variation of tPO2 (SVtPO2). The effects of systemic condition (normoxia/hypoxia) and retinal depth on MtPO2 and SVtPO2 were determined by mixed linear model.

RESULTS: Each 3D retinal tPO2 volume was approximately 500 × 750 × 200 μm (horizontal × vertical × depth) and consisted of 45 en face tPO2 images through the retinal depth. MtPO2 at the chorioretinal interface was significantly correlated with systemic arterial oxygen tension (P = 0.007; N = 9). There were significant effects of both systemic condition and retinal depth on MtPO2 and SVtPO2, such that both were lower under hypoxia than normoxia and higher in the outer retina than inner retina (P < 0.001).

CONCLUSION: For the first time, 3D imaging of retinal tPO2 was demonstrated, with potential future application for assessment of physiological alterations in animal models of retinal diseases.

PMID: 28956656

Retinal pigment epithelium cholesterol efflux mediated by the 18kDa translocator protein, TSPO, a potential target for treating age-related macular degeneration.

Biswa L, Zhou X, Dhillon B, Graham A, Shu X.

Abstract: Cholesterol accumulation beneath the retinal pigment epithelium (RPE) cells is supposed to

Pathogenesis


Retinal pigment epithelium cholesterol efflux mediated by the 18kDa translocator protein, TSPO, a potential target for treating age-related macular degeneration.

Biswa L, Zhou X, Dhillon B, Graham A, Shu X.

Abstract: Cholesterol accumulation beneath the retinal pigment epithelium (RPE) cells is supposed to
contribute the pathogenesis of age-related macular degeneration (AMD). Cholesterol efflux genes (APOE and ABCA1) were identified as risk factors for AMD, although how cholesterol efflux influences accumulation of this lipid in sub-RPE deposits remains elusive. The 18kDa translocator protein, TSPO, is a cholesterol-binding protein implicated in mitochondrial cholesterol transport. Here, we investigate the function of TSPO in cholesterol efflux from the RPE cells. We demonstrate in RPE cells that TSPO specific ligands promote cholesterol efflux to acceptor (apo)lipoprotein and human serum, while loss of TSPO resulted in impaired cholesterol efflux. TSPO/- RPE cells also had significantly increased production of reactive oxygen species (ROS) and upregulated expression of proinflammatory cytokines (IL-1β and TNFα). Cholesterol (oxidized LDL) uptake and accumulation were markedly increased in TSPO/- RPE cells. Finally, in aged RPE cells, TSPO expression was reduced and cholesterol efflux impaired. These findings provide a new pharmacological concept to treat early AMD patients by stimulating cellular cholesterol removal with TSPO specific ligands or by overexpression of TSPO in RPE cells.

PMID: 28973423


ApoA-I Mimetic Peptide 4F Reduces Age-Related Lipid Deposition in Murine Bruch's Membrane and Causes Its Structural Remodeling.


PURPOSE: Accumulation of lipoprotein-derived lipids including esterified and unesterified cholesterol in Bruch's membrane of human eyes is a major age-related change involved in initiating and sustaining soft drusen in age-related macular degeneration (AMD). The apolipoprotein (apo) A-I mimetic peptide 4F is a small anti-inflammatory and anti-atherogenic agent, and potent modifier of plasma membranes. We evaluated the effect of intravitreally-injected 4F on murine Bruch’s membrane.

METHODS: We tested single intravitreal injections of 4F doses (0.6 µg, 1.2 µg, 2.4 µg, and placebo scrambled peptide) in ApoEnull mice ≥10 months of age. After 30 days, mice were euthanized. Eyes were processed for either direct immunofluorescence detection of esterified cholesterol (EC) in Bruch's membrane whole mounts via a perfringolysin O-based marker linked to green fluorescent protein or by transmission electron microscopic visualization of Bruch's membrane integrity. Fluorescein isothiocyanate-conjugated 4F was traced after injection.

RESULTS: All injected eyes showed a dose-dependent reduction of Bruch's membrane EC with a concomitant ultrastructural improvement compared to placebo treated eyes. At a 2.4 µg dose of 4F, EC was reduced on average by ~60% and Bruch's membrane returned to a regular pentalaminar structure and thickness. Tracer studies confirmed that injected 4F reached intraocular targets.

CONCLUSION: We demonstrated a highly effective pharmacological reduction of EC and restoration of Bruch's membrane ultrastructure. The apoA-I mimetic peptide 4F is a novel way to treat a critical AMD disease process and thus represents a new candidate for treating the underlying cause of AMD.

PMID: 28972410


Resveratrol reverses the adverse effects of bevacizumab on cultured ARPE-19 cells.


Abstract: Age-related macular degeneration (AMD) and proliferative diabetic retinopathy (PDR) are one of the major causes of blindness caused by neo-vascular changes in the retina. Intravitreal anti-VEGF
injections are widely used in the treatment of wet-AMD and PDR. A significant percentage of treated patients have complications of repeated injections. Resveratrol (RES) is a polyphenol phytoalexin with antioxidative, anti-inflammatory and anti-proliferative properties. Hence, we hypothesized that if RES is used in combination with bevacizumab (BEV, anti-VEGF), it could reverse the adverse effects that precipitate fibrotic changes, drusen formation, tractional retinal detachment and so on. Human retinal pigment epithelial cells were treated with various combinations of BEV and RES. There was partial reduction in secreted VEGF levels compared to untreated controls. Epithelial-mesenchymal transition was lower in BEV + RES treated cultures compared to BEV treated cultures. The proliferation status was similar in BEV + RES as well as BEV treated cultures both groups. Phagocytosis was enhanced in the presence of BEV + RES compared to BEV. Furthermore, we observed that notch signaling was involved in reversing the adverse effects of BEV. This study paves way for a combinatorial strategy to treat as well as prevent adverse effects of therapy in patients with wet AMD and PDR.

PMID: 28947815


Dynamic thiol/disulfide homeostasis in patients with age-related macular degeneration.
Aktaş S, Sağdık HM, Tetikoğlu M, Aktaş H, Özcura F, Uçar F, Alışık M, Ergin M.

PURPOSE: We evaluated dynamic thiol/disulfide homeostasis (TDH), malondialdehyde (MDA) levels, and catalase (CAT) activity in patients with age-related macular degeneration (AMD). All analyzes were conducted on plasma samples.

METHODS: Thirty-two patients with AMD and 38 age-matched healthy controls were included. Native thiol, total thiol, and disulfide levels and TDH status were determined using a novel, automated assay. MDA levels and CAT activity were determined. Percentages were compared using the chi-squared test. The Student's t-test and Mann-Whitney U-test were used to compare quantitative variables.

RESULTS: Native thiol levels were significantly lower (p=0.004) in patients with AMD (272.02 ± 52.41 µmol/l) than in healthy individuals (307.82 ± 47.18 µmol/l), whereas disulfide levels were significantly higher (p<0.001) in patients with AMD than in controls (21.64 ± 5.59 vs. 14.48 ± 5.37 µmol/L). Dynamic TDH was also significantly lower (p<0.001) in patients with AMD than in controls (13.41 ± 4.3 vs. 25.41 ± 14.52 µmol/l). No significant differences were evident in total thiol or MDA levels. Mean CAT activity was significantly higher (p=0.043) in patients with AMD compared with controls (0.035 vs. 0.018 k/ml).

CONCLUSIONS: The antioxidant/oxidant balance demonstrated by dynamic TDH is shifted to the oxidative side in patients with AMD.

PMID: 28954023

Am J Pathol. 2017 Sep 20. [Epub ahead of print]

Proteolytic Degradation and Inflammation Play Critical Roles in Polypoidal Choroidal Vasculopathy.

Abstract: Polypoidal choroidal vasculopathy (PCV) is a common subtype of wet age-related macular degeneration (AMD) in Asian populations while choroidal neovascularization (CNV) is the typical subtype in Western populations. The etiology of PCV is unknown. By comparing the phenotype of a PCV mouse model expressing protease HTRA1 in retinal pigment epithelium with transgenic mice expressing the inactive HTRA1S328A, we showed that HTRA1 mediated degradation of elastin in choroidal vessels is critical for the development of PCV, which exhibited destructive extracellular matrix remodeling and vascular smooth muscle cell loss. Compared with weak PCV, severe PCV exhibited prominent immune
complex deposition, complement activation, and infiltration of inflammatory cells, suggesting inflammation plays a key role in PCV progression. Importantly, we validated these findings in human PCV specimens. Intravitreal delivery of an HTRA1 inhibitor (DPMFKLboroV) was effective (36% lesion reduction, P = 0.009) in preventing PCV initiation but ineffective in treating existing lesions. Anti-inflammatory glucocorticoid was effective in preventing PCV progression but ineffective in preventing PCV initiation. These results suggest that PCV pathogenesis occurs through two stages. The initiation stage is mediated by proteolytic degradation of extracellular matrix proteins due to increased HTRA1 activity while the progression stage is driven by inflammatory cascades. This study provides a basis for understanding the differences between PCV and CNV, and helps guide the design of effective therapies for PCV.

PMID: 28941979

Semin Immunopathol. 2017 Sep 25. [Epub ahead of print]

The eye as a complement dysregulation hotspot.

Clark SJ, Bishop PN.

Abstract: Complement turnover is tightly regulated throughout the human body in order to prevent over-activation and subsequent damage from inflammation. In the eye, low-level complement activation is maintained to provide immune tolerance in this immune privileged organ. Conversely, the complement system is suppressed in the cornea to protect it from continuous immunological insult. Over-activation of the complement cascade has been implicated in the disease progression of glaucoma and diabetic retinopathy and is now known to be a central driver in the pathogenesis of age-related macular degeneration (AMD). Indeed, it is with AMD where the most recent and exciting work has been carried out with complement-based therapies entering into clinical trials. However, the success of these trials will depend upon delivering the therapeutics to the correct anatomical sites within the eye, so a full understanding of how complement regulation is compartmentalized in the eye is required, a topic that will be highlighted in this review.

PMID: 28948331


Restoring vision in mice with retinal degeneration using multicharacteristic opsin.

Wright W, Gajjeraman S, Batabyal S, Pradhan S, Bhattacharya S, Mahapatra V, Tripathy A, Mohanty S.

Abstract: Retinal degenerative diseases, such as retinitis pigmentosa (RP) and dry age-related macular degeneration, have led to loss of vision in millions of individuals. Currently, no surgical or medical treatment is available, although optogenetic therapies are in clinical development. We demonstrate vision restoration using multicharacteristics opsin (MCO1) in animal models with degenerated retina. MCO1 is reliably delivered to specific retinal cells via intravitreal injection of adeno-associated virus (vMCO1), leading to significant improvement in visually guided behavior conducted using a radial arm water maze. The time to reach the platform and the number of error arms decreased significantly after delivery of MCO1. Notably, the improvement in visually guided behavior was observed even at light intensity levels orders of magnitude lower than that required for channelrhodopsin-2 opsin. Viability of vMCO1-treated retina is not compromised by chronic light exposure. Safe virus-mediated MCO1 delivery has potential for effective gene therapy of diverse retinal degenerations in patients.

PMID: 28948190 PMCID: PMC5603575
Epidemiology


Pattern of Ocular Morbidity in the Elderly Population of Northern India.

Baldev VF, Chopra R, Batra N, Singh S.

INTRODUCTION: The frequency of eye diseases has been suggested to start increasing around 40 years of age, with an even steeper increase beginning around 60 years of age. Health promotions and curative and rehabilitative services for the visually impaired elderly population should therefore be a priority in the coming years especially in low and middle income countries.

AIM: To examine the changing pattern of ocular morbidity in the elderly population of Northern India and to determine the socioeconomic status in relation to ocular morbidities.

MATERIALS AND METHODS: A team from the Department of Ophthalmology and Department of Community Medicine, conducted house visits and did a complete eye examination of 450 elderly subjects. They were selected by systematic random sampling from the data base available in the Department of Community Medicine.

RESULTS: A total of 900 eyes were examined. Visual impairment and blindness was seen in 135 (30%) and 36 (8%) individuals respectively. The most common cause of blindness was cataract, followed by corneal opacity, glaucoma, refractive error, diabetic retinopathy, macular scar, age related macular degeneration, retinal detachment, retinitis pigmentosa. Visual impairment was more in individuals with low socioeconomic status.

CONCLUSION: The results of this study suggest that though cataract remains the main cause of blindness, there is an increase in blindness and visual impairment due to corneal diseases and glaucoma which was not seen earlier. The availability and accessibility to eye care facilities particularly for corneal diseases and glaucoma should be increased to reduce blindness in Northern India.

PMID: 28969173 PMCID: PMC5620814


Lin SY, Lin CL, Chang CH, Wu HC, Lin CH, Kao CH.

BACKGROUND: Prostate cancer (PC) can be related to increased systemic oxidative stress and dihydrotestosterone level, which are also reported to be involved in the pathogenesis of age-related macular degeneration (AMD). We conducted a cohort study to determine whether patients with PC have an increased risk of AMD.

PATIENTS AND METHODS: Data were collected from the Taiwan Longitudinal Health Insurance Database for the 1999-2010 period. The study PC cohort comprised 22,084 patients aged ≥18 years with a first diagnosis of PC. The comparison cohort consisted of age-, occupation-, and urbanization level-matched patients at a ratio of 1:1. The primary outcome was the incidence of AMD, which was evaluated using Kaplan-Meier survival analysis and proportional hazards modeling.

RESULTS: The mean follow-up periods (standard deviation) for the patients with AMD in the age-, occupation-, and urbanization level-matched PC cohort and non-PC cohorts were 4.69 (2.90) and 5.51 (2.82) years. The mean age of the PC cohort was 73.9 years and that of the non-PC cohort was 73.2 years, with approximately 85.9% of the patients aged >65 years. The PC cohort had a higher risk of AMD than did the propensity score-matched non-PC cohort with an adjusted hazard ratio of 1.25 (95% confidence interval, 1.12-1.39). Compared with PC cohort receiving no injection hormone therapy, the PC cohort
receiving injection hormone therapy had a lower risk of AMD (adjusted hazard ratio, 0.56; 95% confidence interval, 0.41-0.76).

CONCLUSION: PC is associated with an increased risk of AMD. Patients with PC receiving injected form of androgen deprivation therapy had a lower risk of AMD than patients with PC not receiving injected form of androgen-deprivation therapy.

PMID: 28961846

Genetics & gene therapy

Ophthalmic Genet. 2017 Sep 26:1-5. [Epub ahead of print]

Evaluation of sFLT1 protein levels in human eyes with the FLT1 rs9943922 polymorphism.


PURPOSE: Age-related macular degeneration (AMD) is a devastating disease characterized by central vision impairment in individuals with advanced age. Neovascular AMD is a form of end-stage disease in which choroidal vessel outgrowth occurs beneath the retina. While many hypotheses have been raised as to what triggers the formation of pathological choroidal neovascular membranes, the exact mechanism for their initiation remains unresolved. Polymorphisms in the FLT1 gene have previously been associated with neovascular AMD risk, including the rs9943922 single nucleotide polymorphism (SNP). Here, we aimed to determine the association between the high-risk FLT1 genotype and FLT1 protein levels in human retina or retinal pigment epithelium (RPE)/choroid tissue.

METHODS: Retina and RPE/choroid tissue from 10 human donor eyes was selected from a collection of eyes genotyped for the rs9943922 SNP. Differences in soluble and membrane bound FLT1 protein levels were assessed for retina versus RPE/choroid donor tissue using ELISA and Western blotting analyses. Genotype-associated changes in FLT1 protein levels were also evaluated.

RESULTS: We found soluble FLT1 levels in the RPE/choroid tissue to be approximately three times higher than that of the retina (p < 0.001), while both samples have similar levels of the membrane bound form. When tissue with the rs9943922 SNP was compared with controls, no significant genotypic differences in FLT1 protein levels were observed.

CONCLUSIONS: Based on these data, we conclude that the rs9943922 SNP in the FLT1 gene does not result in a large difference in FLT1 protein levels, regardless of whether it is the soluble or the membrane bound form.

PMID: 28949775


Long non-coding RNA associated-competing endogenous RNAs are induced by clusterin in retinal pigment epithelial cells.

Ye Z, Li Z, He S.

Abstract: Age related macular degeneration is one of the most common causes of vision loss in the elderly. Long noncoding RNAs (lncRNAs) serve important roles in regulating gene expression by acting as competing endogenous RNAs (ceRNAs). However, the roles of specific lncRNAs and their associated ceRNA function induced by clusterin in cultured retinal pigment epithelial (RPE) cells remain to be fully elucidated. Based on high throughput sequencing data from RPE cells treated with or without clusterin, the present study identified differentially expressed mRNAs, lncRNAs and microRNAs (miRNAs). A lncRNA-mRNA-microRNA (miRNA) network (ceRNA network) was subsequently constructed based on the
bioinformatic database miRanda and miRNA targets database miRTarBase. These results demonstrated the expression pattern of several IncRNAs, and a clear clusterin-associated ceRNA network in RPE cells, which included 75 IncRNAs and 32 miRNAs in RPE cells induced by clusterin. Collectively, the present study uncovered and characterized via bioinformatics the global properties of the ceRNA network in human RPE cells in response to clusterin. These results may aid in the elucidation of the molecular mechanisms of clusterin in age-related macular degeneration.

PMID: 28944909


MMP-2 Rs24386 (C-->T) gene polymorphism and the phenotype of age-related macular degeneration.


AIM: To examine the MMP-2 (-1306 C/T) gene polymorphism and the phenotype characterized by soft and hard drusen of early age-related macular degeneration (AMD) and geographic atrophy of late AMD form.

METHODS: The study enrolled 850 investigations (290 AMD patients with soft and hard drusen, 34 with geographic atrophy and a random sample of the population n=526). Early AMD was classified according to the International Classification and Grading System. For geographic atrophy diagnosis the Age-Related Eye Disease Study classification was used. The potential association with single nucleotide polymorphisms on MMP-2 Rs243865 was evaluated for all patients, adjusted for age and sex. The genotyping test of MMP-2 Rs243865 (C-->T) was conducted using the real-time polymerase chain reaction method.

RESULTS: MMP-2 (-1306 C/T) C/C genotype was more frequently detected in AMD patients with hard drusen than the soft drusen or control group (66.43% vs 53.74%, vs 54.94%, P=0.047). Logistic regression analysis showed that the MMP-2 (-1306) C/C genotype increased the likelihood to develop hard drusen in AMD patients (OR=1.7, 95% CI: 1.06-2.74; P=0.028). No association between MMP-2 (-1306 C/T) gene polymorphism in patients with atrophic AMD and control group was found (54.94%, 37.64%, 7.41% vs 50%, 38.24%, 11.76%; P=0.6).

CONCLUSION: The MMP-2 Rs24386 (C-->T) polymorphism is found to be associated with the development of hard drusen in patients with AMD.

PMID: 28944191 PMCID: PMC5596217

Exp Eye Res. 2017 Sep 21;165:65-77. [Epub ahead of print]

Regulated efflux of photoreceptor outer segment-derived cholesterol by human RPE cells.


Abstract: Genetic studies have linked age-related macular degeneration (AMD) to genes involved in high-density lipoprotein (HDL) metabolism, including ATP-binding cassette transporter A1 (ABCA1). The retinal pigment epithelium (RPE) handles large amounts of lipids, among others cholesterol, partially derived from internalized photoreceptor outer segments (OS) and lipids physiologically accumulate in the aging eye. To analyze the potential function of ABCA1 in the eye, we measured cholesterol efflux, the first step of HDL generation, in RPE cells. We show the expression of selected genes related to HDL metabolism in mouse and human eyecups as well as in ARPE-19 and human primary RPE cells. Immunofluorescence staining revealed localization of ABCA1 on both sides of polarized RPE cells. This was functionally confirmed by directional efflux to apolipoprotein AI (ApoA-I) of 3H-labeled cholesterol given to the cells via serum or via OS. ABCA1 expression and activity was modulated using a liver-X-receptor (LXR) agonist and an ABCA1
neutralizing antibody, demonstrating that the efflux was ABCA1-dependent. We concluded that the ABCA1-mediated lipid efflux pathway, and hence HDL biosynthesis, is functional in RPE cells towards both the basal (choroidal) and apical (subretinal) space. Impaired activity of the pathway might cause age-related perturbations of lipid homeostasis in the outer retina and thus may contribute to disease development and/or progression.

PMID: 28943268

**Stem cells**


Differentiation of RPE cells from integration-free iPS cells and their cell biological characterization.


BACKGROUND: Dysfunction of the retinal pigment epithelium (RPE) is implicated in numerous forms of retinal degeneration. The readily accessible environment of the eye makes it particularly suitable for the transplantation of RPE cells, which can now be derived from autologous induced pluripotent stem cells (iPSCs), to treat retinal degeneration. For RPE transplantation to become feasible in the clinic, patient-specific somatic cells should be reprogrammed to iPSCs without the introduction of reprogramming genes into the genome of the host cell, and then subsequently differentiated into RPE cells that are well characterized for safety and functionality prior to transplantation.

METHODS: We have reprogrammed human dermal fibroblasts to iPSCs using nonintegrating RNA, and differentiated the iPSCs toward an RPE fate (iPSC-RPE), under Good Manufacturing Practice (GMP)-compatible conditions.

RESULTS: Using highly sensitive assays for cell polarity, structure, organelle trafficking, and function, we found that iPSC-RPE cells in culture exhibited key characteristics of native RPE. Importantly, we demonstrate for the first time with any stem cell-derived RPE cell that live cells are able to support dynamic organelle transport. This highly sensitive test is critical for RPE cells intended for transplantation, since defects in intracellular motility have been shown to promote RPE pathogenesis akin to that found in macular degeneration. To test their capabilities for in vivo transplantation, we injected the iPSC-RPE cells into the subretinal space of a mouse model of retinal degeneration, and demonstrated that the transplanted cells are capable of rescuing lost RPE function.

CONCLUSIONS: This report documents the successful generation, under GMP-compatible conditions, of human iPSC-RPE cells that possess specific characteristics of healthy RPE. The report adds to a growing literature on the utility of human iPSC-RPE cells for cell culture investigations on pathogenicity and for therapeutic transplantation, by corroborating findings of others, and providing important new information on essential RPE cell biological properties.

PMID: 28969679


Bioengineered Bruch's-like extracellular matrix promotes retinal pigment epithelial differentiation.

McLenachan S, Hao E, Zhang D, Zhang L, Edel M, Chen F.

Abstract: In the eye, the retinal pigment epithelium (RPE) adheres to a complex protein matrix known as Bruch's membrane (BrM). The aim of this study was to provide enriched conditions for RPE cell culture through the production of a BrM-like matrix. Our hypothesis was that a human RPE cell line would deposit an extracellular matrix (ECM) resembling BrM. The composition and structure of ECM deposited by
ARPE19 cells (ARPE19-ECM) was characterized. To produce ARPE19-ECM, ARPE19 cells were cultured in the presence dextran sulphate. ARPE19-ECM was decellularized using deoxycholate and characterized by immunostaining and western blot analysis. Primary human RPE and induced pluripotent stem cells were seeded onto ARPE19-ECM or geltrex coated surfaces and examined by microscopy or RT-PCR. Culture of ARPE19 cells with dextran sulphate promoted nuclear localization of SOX2, formation of tight junctions and deposition of ECM. ARPE19 cells deposited ECM proteins found in the inner layers of BrM, including fibronectin, vitronectin, collagens IV and V as well as laminin-alpha-5, but not those found in the middle elastic layer (elastin) or the outer layers (collagen VI). ARPE19-ECM promoted pigmentation in human RPE and pluripotent stem cell cultures. Expression of RPE65 was significantly increased on ARPE19-ECM compared with geltrex in differentiating pluripotent stem cell cultures. ARPE19 cells deposit ECM with a composition and structure similar to BrM in the retina. Molecular cues present in ARPE19-ECM promote the acquisition and maintenance of the RPE phenotype. Together, these results demonstrate a simple method for generating a BrM-like surface for enriched RPE cell cultures.

PMID: 28955745 PMCID: PMC5614661

Acta Biomater. 2017 Sep 23. [Epub ahead of print]

Design, Development and Characterization of Synthetic Bruch’s Membranes.


Abstract: Age-related macular degeneration (AMD) is a leading cause of blindness, and dry AMD has no effective treatment. Retinal constructs comprising retinal pigment epithelium (RPE) cells supported by electrospun scaffolds have been investigated to treat dry AMD. However, electrospun scaffolds studied to-date do not mimic the structural microenvironment of human Bruch’s membrane (BM), essential for native-like RPE monolayers. The aim of this study was to develop a structurally biomimetic scaffold designed to support a functional RPE monolayer, comprising porous, electrospun nanofibrous membranes (ENMs), coated with laminin, mimicking the inner collagenous layer (ICL) and basal RPE lamina respectively, the cell supporting layers of the BM. In vitro evaluation showed 70 nm PLLA ENMs adsorbed high amounts of laminin and supported functional RPE monolayers, exhibiting 3D polygonal-cobblestone morphology, apical microvilli, basal infoldings, high transepithelial resistance (TER), phagocytic activity and expression of signature RPE markers. 70 nm PLLA ENMs were successfully implanted into the subretinal space of RCS-ryd+p+/LAV rats, also commonly known as rdy rats. At week 4, in the absence of immunosuppressants, implanted PLLA ENMs were surrounded by a significantly low number of activated microglial cells, compared to week 1, indicating no adverse long-term immune response. In conclusion, we successfully designed and tested ENMs emulating the RPE cell supporting layers of the BM, and found 70 nm PLLA ENMs to be best suited as scaffolds for fabricating retinal constructs.

STATEMENT OF SIGNIFICANCE:

Age related macular degeneration (AMD) is a leading cause of vision loss in the developed world, with an increasing number of people suffering from blindness or severe visual impairment. Transplantation of retinal pigment epithelium (RPE) cells supported on a synthetic, biomimetic-like Bruch’s membrane (BM) is considered a promising treatment. However, the synthetic scaffolds used do not mimic the microenvironment of the RPE cell supporting layers, required for the development of a functional RPE monolayer. This study indicated that porous, laminin coated, 70 nm PLLA ENMs supported functional RPE monolayers, exhibiting 3D polygonal-cobblestone morphology, apical microvilli, basal infoldings, high transepithelial resistance (TER), phagocytic activity and expression of signature RPE markers. These findings indicate the potential clinical use of porous, laminin coated, 70 nm PLLA ENMs in fabricating retinal constructs aimed at treating dry AMD.

PMID: 28951331
Diet, lifestyle & low vision


Effect of Dietary Supplementation With Lutein, Zeaxanthin, and ω-3 on Macular Pigment: A Randomized Clinical Trial.


IMPORTANCE: Nutritional uptake of lutein, zeaxanthin, and ω-3 polyunsaturated fatty acids may increase macular pigment optical density (MPOD) and thereby protect against the development of age-related macular degeneration (AMD).

OBJECTIVES: To estimate the efficiency of dietary supplementation containing lutein, zeaxanthin, ω-3 polyunsaturated fatty acids, and vitamins to increase the density of macular pigment in first-generation offspring of parents with neovascular AMD.

DESIGN, SETTING, AND PARTICIPANTS: This study was a randomized clinical trial (Lutein Influence on Macula of Persons Issued From AMD Parents [LIMPIA]) with a 6-month treatment period, followed by a 6-month follow-up period. Analyses were based on the intent-to-treat principle. The setting was 2 university hospitals in France (at Bordeaux and Dijon) from January 2011 (first participant first visit) to February 2013 (last participant last visit). The analysis was conducted from January to November 2016. Participants were 120 individuals free of any retinal ocular disease. They were first-generation offspring of parents with neovascular AMD.

INTERVENTIONS: Participants were randomized in a 1:1 ratio to receive either 2 daily dietary supplementation capsules or placebo for 6 months.

MAIN OUTCOMES AND MEASURES: The primary assessment criterion was the evolution of MPOD after 6 months of supplementation (value of both eligible eyes) measured using the modified MPD-Visucam 200 (Carl Zeiss Meditec) and the modified Heidelberg Retina Angiograph (Heidelberg Engineering) (HRA) at 0.98° eccentricity. The statistical analysis was adjusted for hospital and for risk factors.

RESULTS: Overall, 120 participants (60 in each group) were included, and 239 eyes were analyzed (119 in the lutein plus zeaxanthin [L + Z] group and 120 in the placebo group). Their mean (SD) age was 56.7 (6.6) years, and 71.7% (n = 86) were female. A statistically significant increase in plasma lutein and zeaxanthin was shown in the L + Z group after 3 months and 6 months of treatment compared with the placebo group. However, the difference between groups in the evolution of MPOD measured by HRA 0.98° eccentricity between 6 months and baseline was 0.036 (95% CI, -0.037 to 0.110) (P = .33).

CONCLUSIONS AND RELEVANCE: Among first-generation offspring of parents with neovascular AMD in the LIMPIA trial, MPOD as measured with the modified HRA and the MPD-Visucam was not modified after 6 months of lutein and zeaxanthin dietary supplementation despite plasma levels showing continuous exposure to lutein and zeaxanthin. Further research is necessary to understand the mechanism of absorption and metabolism of these nutrients in the macula, the best way to measure MPOD, and the clinical benefit for the patients.

PMID: 28973076


The Role of Macular Pigment Density Measures in Future Clinical Studies of Age-Related Macular Degeneration.

Lindblad AS.

PMID: 28973143
Functional changes at the preferred retinal locus in subjects with bilateral central vision loss.

Krishnan AK, Bedell HE.

PURPOSE: Subjects with bilateral central vision loss (CVL) use a retinal region called the preferred retinal locus (PRL) for performing various visual tasks. We probed the fixation PRL in individuals with bilateral macular disease, including age-related macular degeneration (AMD) and Stargardt disease (STGD), for localized sensitivity deficits.

METHODS: Three letter words at the critical print size were presented in the NIDEK MP-1 microperimeter to determine the fixation PRL and its radial retinal eccentricity from the residual fovea in 29 subjects with bilateral CVL. Fixation stability was defined as the median bivariate contour ellipse area (BCEA) from 3 fixation assessments. A standard 10-2 grid (68 locations, 2° apart) was used to determine central retinal sensitivity for Goldmann size II test spots. Baseline and follow-up supra-threshold screening of the fixation PRL for localized sensitivity deficits was performed using high density (0.2° or 0.3° apart) 0 dB Goldmann size II test spots. Custom MATLAB code and a dual bootstrapping algorithm were used to register test-spot locations from the baseline and follow-up tests. Locations where the 0 dB test spots were not seen on either test were labeled as micro-scotomas (MSs).

RESULTS: Median BCEA correlated poorly with the radial eccentricity of the fixation PRL. Mean (±SD) sensitivity around the PRL from 10-2 testing was 4.93 ± 4.73 dB. The average percentage of MSs was similar for patients with AMD (25.4%), STGD (20.3%), and other etiologies of CVL (27.1%).

CONCLUSIONS: The fixation PRL in subjects with bilateral CVL frequently includes local regions of sensitivity loss.

PMID: 28971293

Comparing Analytic Hierarchy Process and Discrete-Choice Experiment to Elicit Patient Preferences for Treatment Characteristics in Age-Related Macular Degeneration.

Danner M, Vennedey V, Hiligsmann M, Fauser S, Gross C, Stock S.

BACKGROUND: In this study, we conducted an analytic hierarchy process (AHP) and a discrete choice experiment (DCE) to elicit the preferences of patients with age-related macular degeneration using identical attributes and levels.

OBJECTIVES: To compare preference-based weights for age-related macular degeneration treatment attributes and levels generated by two elicitation methods. The properties of both methods were assessed, including ease of instrument use.

METHODS: A DCE and an AHP experiment were designed on the basis of five attributes. Preference-based weights were generated using the matrix multiplication method for attributes and levels in AHP and a mixed multinomial logit model for levels in the DCE. Attribute importance was further compared using coefficient (DCE) and weight (AHP) level ranges. The questionnaire difficulty was rated on a qualitative scale. Patients were asked to think aloud while providing their judgments.

RESULTS: AHP and DCE generated similar results regarding levels, stressing a preference for visual improvement, frequent monitoring, on-demand and less frequent injection schemes, approved drugs, and mild side effects. Attribute weights derived on the basis of level ranges led to a ranking that was opposite to the AHP directly calculated attribute weights. For example, visual function ranked first in the AHP and last on the basis of level ranges.

CONCLUSIONS: The results across the methods were similar, with one exception: the directly measured
AHP attribute weights were different from the level-based interpretation of attribute importance in both DCE and AHP. The dependence/independence of attribute importance on level ranges in DCE and AHP, respectively, should be taken into account when choosing a method to support decision making.

PMID: 28964450


Associations between fruits, vegetables, vitamin A, β-carotene and flavonol dietary intake, and age-related macular degeneration in elderly women in Korea: the Fifth Korea National Health and Nutrition Examination Survey.

Kim EK, Kim H, Kwon O, Chang N.

BACKGROUND/OBJECTIVES: Age-related macular degeneration (AMD) is one of the principal causes of blindness. This study investigated the association between diet and the prevalence of AMD in elderly Korean women.

SUBJECTS/METHODS: Study subjects were women aged ≥65 years (n=1008) from the Korea National Health and Nutrition Examination Survey (2010-2012). The presence of early- and late-onset AMD was determined on the basis of a fundus photograph from a health examination survey. Food intake was estimated using 24 h recall.

RESULTS: The prevalence of AMD was 18.8% in elderly women in Korea. Multiple logistic regression analysis showed a significant negative association between vegetable intake and AMD (odds ratio (OR) 0.44, 95% confidence interval (CI) 0.25, 0.77, P for trend=0.002) after adjusting for age, body mass index, postmenopausal period, duration of hormone replacement therapy, residential area, education level, family income, smoking status, alcohol consumption, dietary supplement use and total energy intake. After adjusting for potential confounders, the ORs between extreme quartiles were 0.55 (95% CI 0.29, 1.05, P for trend=0.070) for fruit and vegetable intake, 0.38 (95% CI 0.21, 0.68, P for trend=0.001) for vitamin A, 0.36 (95% CI 0.19, 0.67, P for trend<0.001) for β-carotene and 0.45 (95% CI 0.25, 0.82, P for trend=0.008) for flavonols.

CONCLUSIONS: These results suggest that higher consumption of fruits and vegetables containing antioxidant nutrients and phytochemicals may provide some protection against AMD.

PMID: 28952611

Nutrients. 2017 Sep 29;9(10).

Efficacy of a Fatty Acids Dietary Supplement in a Polyethylene Glycol-Induced Mouse Model of Retinal Degeneration.

Cammalleri M, Dal Monte M, Locri F, Lardner E, Kvanta A, Rusciano D, André H, Bagnoli P.

Abstract: Current knowledge of the benefits of nutrition supplements for eye pathologies is based largely on the use of appropriate animal models, together with defined dietary supplementation. Here, C57BL6 mice were subretinally injected with polyethylene glycol (PEG)-400, an established model of retinal degeneration with a dry age-related macular degeneration (AMD)-like phenotype, an eye pathology that lacks treatment. In response to PEG-400, markers of the complement system, angiogenesis, inflammation, gliosis, and macrophage infiltration were upregulated in both retinas and retinal pigment epithelium (RPE)/choroids, whereas dietary supplementation with a mixture based on fatty acids counteracted their upregulation. Major effects include a reduction of inflammation, in both retinas and RPE/choroids, and an inhibition of macrophage infiltration in the choroid, yet not in the retina, suggesting a targeted action through the choroidal vasculature. Histological analysis revealed a thinning of the outer nuclear layer (ONL), together with dysregulation of the epithelium layer in response to PEG-400. In addition, immunohistofluorescence
demonstrated Müller cell gliosis and macrophage infiltration into subretinal tissues supporting the molecular findings. Reduced ONL thickness, gliosis, and macrophage infiltration were counteracted by the diet supplement. The present data suggest that fatty acids may represent a useful form of diet supplementation to prevent or limit the progression of dry AMD.

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Lipofuscin Formation Catalyzed by the Milk Protein Beta-Lactoglobulin: Lysine Residues in Cycloretinal Synthesis.


Abstract: Lipofuscin is toxic autofluorescent byproducts of the visual cycle. The accumulation of lipofuscin such as cycloretinal in the retina is attributed to playing a role in the progression of age-related macular degeneration (AMD). Intriguingly, the milk protein β-lactoglobulin (BLG) can promote the cyclodimerization of all-trans retinal to cycloretinal both in vitro and in vivo. Here, site-directed mutagenesis of BLG as well as mass spectrometric analysis with substrate analogs demonstrate that lysine residues play a key role in catalysis. It is also shown that catalytic activity necessitates the presence of a physical binding site and cannot be mediated by a peptide chain. These studies provide insight on the mechanism of the cyclodimerization process and provide a model system for biocatalysis and biosynthesis of cycloretinal in vivo. In the long term, these studies may pave the way for drug development and inhibitor design as an early treatment regimen for AMD.

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Epilutein for Early-Stage Age-Related Macular Degeneration: A Randomized and Prospective Study.

Forte R, Panzella L, Cesarano I, Cennamo G, Eidenberger T, Napolitano A.

PURPOSE: The hypothesis that oral supplementation of the epilutein/lutein combination could augment the macular pigment optical density (MPOD) in patients with age-related macular degeneration (AMD) was tested.

METHODS: In a prospective randomized interventional study, 40 consecutive patients with early-stage AMD were recruited. After a 2-week run-in period, patients were randomly treated with a daily oral administration of 8 mg epilutein and 2 mg lutein (group 1) or 10 mg lutein (group 2) for 2 months. At baseline (BL) and 1-month (M1) and 2-month visits (M2), all patients underwent a complete ophthalmological examination, including measurement of MPOD in a 7° area (Visucam 200; Carl Zeiss Meditec, Milan, Italy). Xanthophylls were quantified in plasma, as well as the HDL, non-HDL, and erythrocyte fractions at each study visit.

RESULTS: Twenty-one patients (mean age 69.4 ± 6.7 years, 35 eyes) were included in group 1. Mean MPOD was 0.203 ± 0.02 optical density units (ODU) at BL, and increased to 0.214 ± 0.04 ODU at M1 (p = 0.008) and 0.206 ± 0.03 ODU at M2 (p = 0.04). Sixteen patients (mean age 72.0 ± 6.3 years, 29 eyes) were included in group 2. Mean MPOD was 0.215 ± 0.03 at BL, which reduced to 0.202 ± 0.03 ODU at M1 (p = 0.003) and 0.207 ± 0.02 ODU at M2 (p < 0.001). A rise in the systemic level of total xanthophylls was observed at M1 for both groups. At M2, total xanthophylls were significantly increased only in group 1 and decreased in group 2.

CONCLUSION: In patients with early-stage AMD, the administration of lutein in combination with epilutein was associated with an increased MPOD compared to the administration of lutein alone.

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Evaluation of a clinical decision-making aid for nutrition advice in age-related macular degeneration.

Stevens R, Bartlett H, Cooke R.

Abstract: Age-related macular disease (AMD) is a multifactorial degenerative condition affecting the central area of the retina. Patients with AMD report that eye care practitioners are not giving consistent advice regarding nutrition and reported confusion as to what advice, if any, to follow. The aim of this study was to design and conduct a preliminary evaluation of a flowchart to support eye care practitioners in providing accurate, evidence-based nutritional advice to their patients. A flowchart was designed to take practitioners through a decision-making process that would determine whether a patient matched the Age-Related Eye Disease Study (AREDS) 2 eligibility criteria for supplementation. The flowchart was evaluated using a qualified and student optometrist cohort, with both cohorts completing confidence scales and students completing clinical scenarios. Qualified participants showed a significant increase in confidence scores from the initial survey (M = 69.7%, standard deviation [SD] = 16.2%) to the second survey after use of the flowchart for 2 weeks (M = 82.1%, SD = 11.6%; t(45) = 7.33, p < .001; rs = .61, p < .001). The student participants also increased confidence scored after receiving the flowchart (M of first survey = 41.7, SD = 14.6; M of second survey = 69.1, SD = 1.7; t(25) = 7.92, d = .81, p < .001) and increased the number of correct answers on five clinical scenarios. Overall, the flowchart has proved to be useful in boosting the self-efficacy of both qualified practitioners and student practitioners, as well as improving clinical decisions made by student practitioners.

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