Melkonyan, Tobias Stupp, Solon Thanos. Germany; 3Excellence Cluster “Cells in Motion”, Westfalian-Wilhelms-University Muenster, Muenster, Boston, MA; 2Institute for Experimental Ophthalmology, School of Medicine, Westfalian-Wilhelms-University Muenster, Muenster, Germany; 3Institute for Experimental Ophthalmology, School of Medicine, Westfalian-Wilhelms-University Muenster, Muenster, Germany.

**Purpose:** Amblyopia is a developmental disease characterized by impaired visual acuity. The underlying molecular pathomechanisms remain unclear. The present work was undertaken to examine the retinal transcripts associated with amblyopia in rats with experimentally induced unilateral amblyopia.

**Methods:** Surgical tarsorrhaphy was performed in the eye lids of newborn Sprague-Dawley (SD) rats prior to eye opening at postnatal day 14, with the contralateral eye serving as the corresponding control. This condition was maintained for over 2 months, after which electroretinograms (ERGs) were recorded and the retinal ganglion cell (RGC) arrangement and number were determined using neuroanatomical tracing. Retinal transcripts were studied using microarray analysis. Regulated mRNAs were confirmed with quantitative reverse-transcriptase PCR, and selected proteins were studied using immunohistochemistry, immunoblots and functional tests in cell cultures.

**Results:** In the eyes deprived of visual experience, we found attenuated ERGs indicating altered physiological neurotransmission. Retrograde neuroanatomical staining disclosed a significantly higher number of RGCs as well as a multilayered distribution of RGCs within the retina on the visually deprived eye, in comparison to non-deprived samples, indicating incomplete programmed cell death and incomplete post-mitotic migration to the ganglion cell layer. At the molecular level, several transcripts were either up- or down-regulated, mostly in association with retinal differentiation, indicating that postnatal differentiation of the retina requires photic stimulation. Most of the transcripts, and in particular FGF-2, could be validated at the mRNA level, while some of the proteins encoded were stainable in retinal sections and gels. When added to primary retinal cell cultures at postnatal stages, FGF-2 stimulated differentiation of ganglion cells, indicating its importance for postnatal retinal development.

**Conclusions:** These data suggest that visual experience crucially shapes the postnatal retinal differentiation, whereas visual deprivation induces changes at both the cellular and molecular levels within the retina, including regulation of powerful differentiation factors such as FGF-2.

**Commercial Relationships:** Melissa Meyer zu Horste, None; Verena Prokosch-Willing, None; Harutyun Melkonyan, None; Tobias Stupp, None; Solon Thanos, None

**Support:** Excellence Cluster “Cells in Motion”, Westfalian-Wilhelms-University Muenster

---

**Fixation Stability and Fixational Eye Movements in Amblyopia**

**Program Number:** 813

**Presentation Time:** 3:45 PM–4:00 PM

**Purpose:** Fixation instability is a limiting factor on acuity in amblyopia. The greater fixation instability cannot be explained by the slow drifts of fixational eye movements. Instead, it is likely that the larger amplitude of fixational saccades contributed to the increased fixation instability in amblyopia.

**Commercial Relationships:** Kristina Irisch, Robert B. Guary, Jing Tian, David L. Guyton, Boris I. Gramatikov, Howard S. Ying. The Wilmer Eye Institute, The Johns Hopkins University School of Medicine, Baltimore, MD.

**Purpose:** To investigate whether detection of interocular fixation instability may serve as a single sensitive test for amblyopia.

**Methods:** Binocular eye movements were recorded at 500 Hz using the EyeLink 1000 eye tracker (SR Research Ltd., Ontario, Canada) and analyzed using EyeLink software and Matlab (Mathworks, Natick, MS). Eight subjects (three non-amblyopic and five amblyopic including one successfully-treated ambylopie; age: 7-44 years)
were asked to fixate on a cross subtending 0.5° at a distance of 57 cm. Intercocular fixation stability was quantified by calculating the minimum-area bivariate contour ellipse (BCEA) encompassing 68% of the difference between right and left eye fixation points during a 20-second binocular viewing epoch. For statistical analysis, BCEA values were normalized by a common logarithm transformation.

**Results:** The amblyopic subjects with persistent vision loss (one anisometropic, two strabismic, and one deprivation; uncorrected visual acuity range 20/60-20/300) showed significantly higher interocular fixation instability (larger 68% BCEAs) than the non-amblyopic subjects (uncorrected visual acuity range 20/20-20/800), and the successfully-treated strabismic amblyope (to the 20/20 level of visual acuity); p < 0.01.

**Conclusions:** Based on our results, interocular fixation instability differentiates amblyopic from non-amblyopic subjects and improves after successful treatment. Intercocular fixation instability may therefore prove to be a single sensitive test for the presence of amblyopia. As a difference measure, it is less susceptible to head motion and calibration error, as well as to conjugate eye motion, and as such is expected to be somewhat immune to latent nystagmus. Intercocular fixation instability may also be used to guide treatment, especially in preverbal children and to assess the efficacy of novel treatments. Further research is required to establish optimal interocular fixation instability thresholds and to determine how specific this measure is to amblyopia.

**Commercial Relationships:** Kristina Irsch, pending (P); Robert B. Geary, pending (P); Jing Tian, pending (P); David L. Guyton, pending (P); Boris I. Gramatikov, pending (P); Howard S. Ying, pending (P)

**Program Number:** 814

**Presentation Time:** 4:00 PM–4:15 PM

**Unmasking potential brain plasticity in amblyopic vision**

Zhong-Lin Lu1, Jiawei Zhou2, Lin Li3, Pan Zhang2, Jie Xi2, Yifeng Zhou2, Chang-Bing Huang2

**Support:** RPB Disney Award (H.S. Ying)

**Program Number:** 815

**Presentation Time:** 4:15 PM–4:30 PM

**Initial Evaluation of the Novel EFG Therapy for the Treatment of Amblyopia**

Puenasanta A. Vera-Diaz1, Gayathri Srinivasan1, Catherine Johnson1, Eric Hussey2, David Spivey3, William Gleason4, Paulette Tattersall5, Bruce D. Moore4, New Engand College of Optometry, Boston, MA; 2Private Practice, Spokane, WA; 3Private Practice, Ft. Worth, TX; 5Foresight Regulatory Strategies, Inc., Wilmington, MA

**Purpose:** Amblyopia is a neurological development disorder that presents with deficits in spatiotemporal vision processing resulting from an active suppression process. The current standard of care for amblyopia involves visual penalization (using patching, often for several hours a day, or atropine) of the “good”, non-amblyopic, eye. We propose and evaluate an alternative treatment method that does not penalize the patient’s ability to see while being treated and promotes normal binocular vision: the Eyetronix Flicker Glasses (EFG). The objective of this study is to evaluate the feasibility and efficacy of the novel EFG Therapy for amblyopia.

**Methods:** 20 children (ages 6-17 years) participated in this initial open-label, multi-center study. Inclusion criteria included: mild to moderate anisometropic amblyopia [difference in logMAR best-corrected visual acuity (BCVA) of 0.2 logMAR (2 lines) or more between the amblyopic and fellow eye]; amblyopic eye BCVA +0.2 to +0.7 logMAR; anisometropia of >1.00DS or >1.50DC; full-time wear of glasses with best-correction for at least 8 weeks prior to the EFG Therapy. EFG is a spectacle frame with liquid crystal lenses and an electronic shutter that allows accurate and rapid alternating rate of occlusion. For this study the EFG were preprogrammed to 7Hz, 50% duty cycle. Subjects were instructed to wear the EFG daily for 1-2 hours during near vision activities. The primary outcome measure was the change of logMAR VA in the amblyopic eye between the dispensing and 3-month visit. Secondary outcome measures included changes in stereopsis and fusion.

**Results:** All but one subject showed improved VA in the amblyopic eye (Mean group improvement -0.09 ± 0.10 logMAR, 1 line). This improvement was significantly larger (p=0.04) than the variation in VA in the non-amblyopic eye. All but two subjects improved MacLeod, 2001). What is surprising is that adapting to gratings of unresolvable spatial frequencies still yielded considerable TAE with an average effect size of 0.48 ± 0.07. Although the TAE thresholds at unresolvable spatial frequencies were smaller than those at the resolvable spatial frequencies (t(25)=5.35, p < 0.01), all the TAE thresholds were significantly greater than 0 (p<0.05). Averaged across observers, the ratio of the cutoff spatial frequencies in the TAE and traditional orientation identification tasks is 1.53 (± 0.08).

**Conclusions:** Neural connections in the amblyopic cortex, at least in V1, may have profoundly developed to represent high spatial frequency information. The demonstration of extant neural connections for high spatial frequencies therefore unmask improvement potentials and provides a strong theoretical basis for developing new therapies for amblyopia. Our paradigm may also serve as a non-invasive probe to diagnose the status of neural connections in other clinical conditions.

**Commercial Relationships:** Zhong-Lin Lu, None; Jiawei Zhou, None; Lin Li, None; Pan Zhang, None; Jie Xi, None; Yifeng Zhou, None; Chang-Bing Huang, None

**Support:** Supported by the Knowledge Innovation Program of the Chinese Academy of Sciences (Y3CX102003), Institute of Psychology, CAS (Y1CX201006) and National Natural Science Foundation of China (NSFC 31230032) to Chang-Bing Huang, and NEI (EY021553) to Zhong-Lin Lu.

©2014, Copyright by the Association for Research in Vision and Ophthalmology, Inc., all rights reserved. Go to iovs.org to access the version of record. For permission to reproduce any abstract, contact the ARVO Office at pubs@arvo.org.
Binocular iPad treatment of amblyopia leads to lasting improvement of visual acuity

Simone L. Li1, Reed Jost1, Sarah Morale1, David Stager2, Lori Dao3, David Stager4, Jr, Eileen Birch1, 4. Crystal Charity Ball Pediatric Vision Evaluation Center, Retina Foundation of the Southwest, Dallas, TX; 2Pediatric Ophthalmology & Center for Adult Strabismus, Dallas, TX; 3Pediatric Ophthalmology & Adult Strabismus, Plano, TX; 4Department of Ophthalmology, University of Texas Southwestern Medical Center, Dallas, TX.

Purpose: Traditional amblyopia treatments (patching/penalization) do not always restore 20/20 vision and the recurrence rate is high within 6 months after the cessation of traditional treatment. We and others recently demonstrated the benefit of binocular treatment for amblyopia in children (Li et al ARVO 2013) and adults (Hess et al 2012, 2013). However, no data to date have determined the durability of the visual acuity improvements as a result of binocular treatment. In this study, we examined whether visual acuity improvements obtained with 4-8 weeks of binocular amblyopia treatment were maintained at 3 and 6 months after the cessation of treatment.

Methods: We assigned 11 amblyopic children to binocular iPad treatment (5-12y). All children had been wearing glasses (if applicable) for at least 3 months and had stable best-corrected visual acuity (BCVA) prior to baseline. All were instructed to play the dichoptic iPad game apps for 4 h/week for 4 weeks and 8 children continued to play the games for an additional 4 weeks. BCVA was measured at baseline, at the 4- and 8-week outcome visits, and at 3 and 6 months after the cessation of treatment. None of the 11 children patched after the cessation of treatment.

Results: At baseline, mean BCVA -se was 0.45±0.07 logMAR (N=11; range: 0.20-0.80 logMAR). With intent-to-treat analysis, BCVA improved significantly with 4-8 weeks of binocular treatment to 0.35±0.07 logMAR (N=11; p<0.01; range: 0.00 to 0.70 logMAR). BCVA improvement gained during the binocular iPad treatment was maintained at 3 months post-treatment (0.37±0.08 logMAR; N=9) and at 6 months post-treatment (0.38±0.08 logMAR; N=9). Among all children who returned for the 3- and/or 6-month post-treatment visits, only one experienced a BCVA regression (≥0.2 logMAR), likely due to measurement error because her BCVA improved by 0.1 logMAR at the 6-month post-treatment visit.

Conclusions: Binocular iPad treatment for amblyopia yielded a significant improvement in BCVA after 4-8 weeks. The obtained improvements in BCVA were maintained. Therefore, binocular iPad treatment is a promising new approach for the treatment of amblyopia.