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The Influence of Cataract on Fluorescence Lifetime Imaging Ophthalmoscopy (FLIO)

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Purpose: To investigate the influence of lens opacifications on fluorescence lifetime imaging ophthalmoscopy (FLIO).

Methods: Forty-seven eyes of 45 patients were included. Mean fluorescence lifetimes (*Tm*) were recorded with a fluorescence lifetime imaging ophthalmoscope in a short spectral channel (SSC) and a long spectral channel (LSC). Retinal and lens autofluorescence lifetimes were measured in subjects before and after cataract surgery. Lens opacification was graded using the Lens Opacities Classification System III (LOCS III) classification.

Results: The retinal *Tm* decreased significantly after cataract surgery in both spectral channels (SSC: -53%, P < 0.0001; LSC: -26%, P = 0.0041). The lens *Tm* differed significantly between the crystalline and the artificial lens in both spectral channels (P < 0.0001). The "nuclear opacity" and "nuclear color" score of the LOCS III classification correlated significantly with the mean *Tm* difference in both spectral channels (P < 0.0001).

Conclusions: Lens opacification results in significantly longer retinal *Tm*. Therefore the lens status has to be considered when performing cross-sectional fluorescence lifetime analysis. Cataract-formation and cataract-surgery needs to be considered when conducting longitudinal studies. Grading of nuclear opacity following the LOCS III classification provides an approximate conversion formula for the mean change of lifetimes, which can be helpful in the interpretation of data in patients with lens opacities.

Translational Relevance: FLIO is significantly influenced by lens opacities. Using a lens opacity grading scheme and measuring fluorescence lifetimes before and after cataract surgery, an approximative conversion formula can be calculated, which enables the comparison of lifetimes after cataract surgery or over the course of time.

Introduction

Fluorescence lifetime imaging ophthalmoscopy (FLIO) is a relatively novel retinal imaging technique.^{1–3} After excitation with a laser and by measuring the emitted autofluorescence of known fluorophores, conclusions on the function and composition of the retinal tissue can be drawn.^{2,4,5} Recent studies showed evidence that FLIO might offer early hints for retinal changes associated with developing retinal pathologies.^{6–9} However, FLIO images can be significantly influenced by optical media opacities such as cataracts. The crystalline lens possesses

a relatively long lifetime compared with an artificial lens, which exhibits no fluorescence.¹⁰ This can be a problem when comparing mixed groups with phakic and pseudophakic eyes and necessitates subgroup analyses.¹¹ After cataract surgery, lifetime measurements might change dramatically, making it difficult for prospective observation and assessment, especially when studying cataract-associated diseases such as retinal detachment.¹² A normalization factor correcting for influences of lens opacities on FLIO would facilitate assessment of fluorescence lifetimes in patients with crystalline lens opacities.

We therefore classified the grade of cataract and measured fluorescence lifetimes of the retina, as well

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	Mean	Range	Standard Deviation
Age (years)	66	25–84	12.4
BCVA presurgery (logMAR Snellen equivalent)	0.33 20/43	-0.1 to 1.4 20/16 - 20/502	0.30 20/40
BCVA postsurgery (logMAR Snellen equivalent)	0.10 20/25	-0.1 to 1.4 20/16 - 20/502	0.27 20/37
LOCS III [*] scores:			
NO	3.3	1.2–6.2	1.24
NC	3.4	0.5–6.9	1.41
C	2.6	0.2–5.8	1.13
P	2.2	0–5.5	1.72

 Table 1.
 Clinical Characteristics of the Enrolled Patients Including Average Lens Grading Scores

^{*}LOCS III: Lens Opacities Classification System III.¹⁶

as from the crystalline and artificial lens, before and after cataract surgery, respectively. The goal was to detect and measure the change in fluorescence lifetimes and create a conversion formula for adjusting for lens opacities in measured retinal FLIO lifetimes.

Methods

This prospective study (ClinicalTrials.gov under NCT01981148) was approved by the local ethics committee and the procedures followed the tenets of the Declarations of Helsinki and the International Ethical Guidelines for Biomedical Research involving Human Subjects. Written informed consent was obtained from all patients before study enrollment.

Patients and Examinations

Patients planned for elective cataract surgery with varying media opacities were recruited at the outpatient department of ophthalmology at the University Hospital of Bern, Switzerland. Patients with media opacities other than cataract were excluded. Patients with retinal pathologies leading to shortterm changes (changes occurring in the timeframe of few weeks) in measured fluorescence lifetimes (such as active choroidal neovascularization and resulting macular edema) were excluded. Patients with mild, static, nonchanging retinal abnormalities (such as drusen in age-dependent macular degeneration) were not excluded, as these pathologies should not have an influence on intraindividual-measured differences before and after surgery.

All measurements (best corrected visual acuity [BCVA], FLIO, macular pigment optical density (MPOD), and slit-lamp photographs) were taken directly before surgery. To reduce acquisition time and potential artefacts,¹³ all pupils were dilated with 0.5%

tropicamide and 2.5% phenylephrine HCl. One week after surgery measurements were repeated. Before the second FLIO measurement the absence of postoperative media opacities at the slit-lamp examination was verified. (Table 1).

FLIO Measurement

Mean fluorescence lifetimes (*Tm*) of the retina and the crystalline and artificial lens were measured using a prototype fluorescence lifetime imaging ophthalmoscope (FLIO) device from Heidelberg Engineering (Heidelberg, Germany).

The FLIO device radiates with a 473-nm pulsed blue laser at 80-MHz repetition rate for excitation of retinal autofluorescence. The emitted fluorescence is registered by time-correlated single-photon counting modules using two highly sensitive hybrid photoncounting detectors (Becker & Hickl, Berlin, Germany). Following detection channels with distinct wavelengths ranges were used: a short spectral channel (SSC, 498–560 nm) and a long spectral channel (LSC, 560– 720 nm). A confocal high-contrast infrared image was recorded simultaneously to ensure that each recorded photon was localized at the correct spatial location. A photon count of 1000 photons per pixel was acquired at least for every location within the image. FLIO imaging took approximately 1.5 to 3 minutes per eye. FLIO images of the lens were recorded with a fixed value of 20 diopters, and the distance to the eye was chosen accordingly to get a sharp delineation of the pupillary border of the iris.

Analysis of Fluorescence Lifetime Data

The Becker & Hickl software (SPCImage 4.4.2) was used for the analysis of recorded lifetime data. A biexponential decay model was applied using a binning factor of one. This procedure resulted in a short (T1)



Figure 1. Representative illustration of the ETDRS grid on a fluorescence lifetime image. It is used to subdivide the lifetime measurements into a central, inner, and outer ring area. The picture on the left represents a fluorescence lifetime image of the SSC of an eye with a cataractous (opacified) lens, and the image on the right the same eye after cataract surgery. The deeper orange-red tones on the right image indicate the generally shorter lifetimes compared with the image on the left, imaged through the cataract. The mean differences in fluorescence lifetimes before and after cataract surgery in the individual ETDRS areas are listed in Table 2.

and a long (T2) lifetime component with corresponding amplitudes a1 and a2.

For topographical mapping and quantitative analysis of measured fluorescence lifetimes, the software "FLIO Reader" (ARTORG Center for Biomedical Engineering Research, University of Bern, Bern, Switzerland) was used. A standardized Early Treatment Diabetic Retinopathy Study (ETDRS¹⁴) grid composed of a center (d = 1 mm), inner (d = 1 mm)3 mm), and outer ring (d = 6 mm) was overlaid over a grayscale-coded lifetime map. The individual FLIO parameters described earlier are displayed for the individual ETDRS grid segments (Fig. 1). For lens images, the region of interest was adjusted to the size of the dilated pupil and the complete visible surface area of the lens was measured (Fig. 2, far right). Tm as well as the individual components T1 and T2 and amplitudes al and a2 were recorded for each of the subdivided sections of the ETDRS grid. Tm represents an amplitude-weighted average of the decay parameters T1 and T2. Reproducibility analysis was not performed because repeatability of FLIO measurements was reported to be very high.¹⁵

Lens Opacity Grading

Lens opacification of the crystalline lens was graded using the Lens Opacities Classification System III (LOCS III) classification¹⁶ on the basis of slit-lamp photographs. Photographs were taken on a Haag-Streit Slit Lamp Model BX 900 LED (Haag-Streit AG, Köniz, Bern, Switzerland) with a Canon EOS



Figure 2. Graph depicting the differences in lens lifetimes. Quantification of mean lens fluorescence lifetimes (lens *Tm*) in ps before (phakic or crystalline lens) and after cataract surgery (pseudophakic or artificial lens) in the SSC (two left columns) and the LSC (two right columns and two FLIO images). On the far right, two representative FLIO images of the lens in the LSC are depicted. The top image represents a phakic eye, the bottom image depicts the same eye, after cataract surgery. The pupillary area, which was used to calculate the mean lens *Tm* is circled in both images.

7D DSLR Camera (Canon Inc., Ota City, Tokyo, Japan) and a Haag-Streit Flash-Unit FU-01 (Haag-Streit AG). Camera settings were set to 1/125 seconds shutter time and ISO400 sensitivity. Slit-beam width was set to the smallest width allowing for adequate illumination. Flash intensity was set to 50%. Aperture size was set to 1.

MPOD Measurement

MPOD was measured using dual wavelength autofluorescence imaging (Heidelberg MultiColor Spectralis, Heidelberg Engineering). The outer reference point was set to 9° eccentricity from the fovea. With this setting, the measurement of reliable macular pigment data has been documented.^{17,18}

Statistical Analysis

For statistical analysis, Prism 8 (GraphPad Software Inc., La Jolla, CA) was used. Data were analyzed for normality using the d'Agostino-Pearson omnibus K2 test. For data following a Gaussian distribution, 1-way analysis of variance with Tukey's multiple comparison test and Pearson correlation was used. For data violating normality, the Friedman Test with Dunn's multiple comparison test and Spearman correlation was used. *Tm* of the lens, subscores NO, NC, and C of the LOCS III grading scheme, and MPOD at 0.5° and 2° followed a Gaussian distribution. *Tm* of the retina, subscore

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P of the LOCS III grading scheme, and MPOD at 9° did not follow a Gaussian distribution. P values of ≤ 0.05 were considered as statistically significant.

Results

A total of 47 eyes of 45 patients (mean age 66, SD ± 12 years, range: 25–84 years) were analyzed. Twentynine patients were men and 16 were women. The most commonly implanted lens type was Hoya XY1 (Hoya Medical, Chromos, Singapore) (n = 31) followed by Tecnis PCB00 (n = 8), and ICB00 (n = 2) (Johnson & Johnson Vision, Jacksonville, FL), Lentis LU-814 VR (Oculentis B.V., LZ Eerbeek, Netherlands) (n = 2), Ankoris Toric (Alyko Medical AB, Bjärred, Sweden) (n = 2), ACRYSof IQ (Alcon, Fort Worth, TX) (n = 1), and ZCT225 (Johnson & Johnson Vision) (n = 1). The mean change in BCVA after surgery was -0.23 logMAR (~20/34 Snellen equivalent, standard error of mean [SEM] = 0.032 logMAR).

FLIO Data of the Retina

Tm of the retina decreased significantly in all areas of the ETDRS grid after cataract surgery in both

spectral channels. The difference between the fluorescence lifetimes before and after cataract surgery was more pronounced in the SSC (Figs. 3, 4). In the total area of the ETDRS grid, Tm (mean \pm SEM) decreased from 716 \pm 54 picoseconds (ps) to 336 \pm 15 ps in the SSC (-53%, P < 0.0001), and from 466 ±26 ps to 347 ± 12 ps in the LSC (-26%, P = 0.0041). For each of the subdivisions of the ETDRS grid (center, inner, and outer ring), the mean differences proved to be statistically significant in both spectral channels, however, it decreased toward the retinal periphery, especially in the LSC (SSC P < 0.0001 for center, inner, and outer circle; LSC P value ranging from < 0.0001 for the center, P =0.0001 for the inner ring, and P = 0.0256 for the outer ring). The detailed changes in lifetime measurements are depicted in Table 2. Retinal fluorescence lifetimes of pseudophakic eyes correlated significantly with age (SSC P = 0.0148, r = 0.36 and LSC P < 0.0001, r = 0.56).

FLIO Data of the Lens

FLIO images of the lens were acquired and analyzed in 38 eyes. The Tm of the lens (mean \pm SEM) differed significantly between the crystalline and the artificial lens in both spectral channels, with a mean



Figure 3. Slit lamp photographs of the lens and corresponding lens and retinal fluorescence lifetime images. Images used to grade the NO subscore of the LOCS III cataract grading scheme are displayed in the top row with a very mild opacified lens (NO score of 1.2, left), moderate opacified lens (NO score of 3.2, middle), and severe opacified lens (NO score of 5.6, right), and corresponding FLIO images of the lens (middle row) and retina (bottom row), whereby the left image represents the SSC and the right image the LSC.



Figure 4. Representative FLIO images of the retina before and after cataract surgery. On the left images of a patient with little lens opacification (NO score 2, A) and on the right with progressed lens opacification (NO score 4.2, B). *Tm* in the center area. Underneath the images, two graphs illustrate the quantification of *Tm* (on the y-axis for both graphs) for the individual ETDRS grid areas: center, inner, and outer ring before (phakic, left column of same color) and after cataract surgery (pseudophakic, right column of same color). The graph on the left illustrates the changes in the SSC and the one on the right the changes in the LSC. An illustration of the ETDRS grid is provided in Figure 1.

	Retinal Lifetime (<i>Tm</i>) in Phakic Eye (±SEM)	Retinal Lifetime (<i>Tm</i>) in Pseudophakic Eye (±SEM)	Mean Reduction (in %)
SSC:			
Center	741 (± 70)	253 (±21)	-488 (66%)
Inner	755 (\pm 60)	332 (± 18)	-423 (56%)
Outer	704 (± 52)	340 (± 14)	-364 (52%)
LSC:			
Center	530 (\pm 52)	297 (\pm 15)	-233 (44%)
Inner	$480(\pm30)$	345 (± 14)	–135 (28%)
Outer	459 (\pm 24)	350 (±11)	-109 (24%

Table 2. Retinal Fluorescence Lifetimes (Tm) in ps Before and After Cataract Surgery Subdivided Into the ETDRS*

*ETDRS: Early Treatment Diabetic Retinopathy Study.¹⁴

Grid areas: center, inner, and outer ring for the SSC (498-560 nm) and the LSC (560-720 nm). A graphic Illustration of the ETDRS grid subdivisions are depicted in Figure 1.

decrease after cataract surgery of -1571 ± 57 ps (P < 0.0001) for the SSC and -1647 ± 58 ps (P < 0.0001) for the LSC. The *Tm* of crystalline lenses was 2742 ±54 ps (in the SSC) and 2361 ±45 ps (in the LSC), and the *Tm* of artificial lenses was 1172 ±28 ps (in the SSC) and 714 ±38 ps (in the LSC) (Fig. 2). A significant correlation of crystalline lense *Tm* with the reduction in retinal *Tm* after cataract surgery could be observed for both spectral channels (P = 0.003 for the SSC and P = 0.002 for the LSC).

LOCS III Grading Data

Slit-lamp photographs of 39 eyes were acquired and graded. Descriptive statistics of the four subscores are included in Table 1. There was a significant correlation with age for nuclear opacity (NO; P = 0.0021), nuclear color (NC; P = 0.0006), cortical cataract (C; P = 0.0017), and posterior subcapsular cataract (P; P = 0.0284). The subscores NO and NC correlated significantly with the corresponding mean change of retinal fluorescence lifetimes in the center, inner,



Figure 5. Obtaining a conversion equation for fluorescence lifetimes in eyes with cataract. Correlation of the NO score of the LOCS III cataract grading scheme (on the x-axis in both graphs) with the mean change in retinal fluorescence lifetimes (*Tm*) in ps after cataract surgery (on the y-axis in both graphs). The graph on the left depicts the SSC and the graph on the right the LSC. An equation to calculate true fluorescence lifetimes in the retina in the presence of advanced lens opacifications was obtained.

and outer ring area of the ETDRS grid in both spectral channels (P < 0.0001 in both spectral channels) (Fig. 5). The cortex (C) score did not show a significant correlation with the fluorescence lifetime change after cataract surgery with P values of 0.2 for the SSC and 0.12 for the LSC. The P score correlated weakly in the inner and outer ring area in both spectral channels (SSC: P = 0.047 for the inner and P = 0.031 for the outer; LSC: P = 0.043 for the inner and P = 0.033 for the outer). In the central area there was no significant correlation found (P = 0.061 and P = 0.08 for the SSC and LSC, respectively). Figure 5 shows the correlation of NO with the mean decrease in retinal fluorescence lifetimes after surgery.

MPOD Data

The MPOD measurements of 26 eyes reached adequate quality to allow for pigment density calculations. The mean macula pigment volume at 0.5° changed from 200 \pm 14 (mean \pm SEM) before surgery to 269 \pm 20 area under the curve after surgery (+35%, P = 0.0001). At 2° eccentricity, it changed from 1858 ± 154 to 2249 ± 212 (+21%, P = 0.003) and at 9° it changed from 7760 ± 835 to 8871 ± 889 (+14%, P = 0.013). When correlating MPOD measurements before cataract surgery with measurements of the same eyes after cataract surgery a significant linear correlation was found (for 0.5° : $r^2 = 0.58$, P < 0.0001; for 2° : r^2 = 0.78, P < 0.0001; for 9°: $r^2 = 0.84, P < 0.0001$). The mean change in MPOD after surgery plotted against the mean change in retinal fluorescence lifetimes in the total ETDRS grid area showed significant correlations for MPOD at 0.5° and 2° in both spectral channels (SSC: P = 0.0073 for 0.5° and P = 0.0129 for 2°;

LSC: P = 0.0002 for 0.5° and P = 0.0003 for 2°), meaning the higher the change in retinal fluorescence lifetimes after surgery, the higher the positive change in measured optical density was observed. The correlation with MPOD at 9° showed no statistical significance.

Discussion

This study provides detailed analysis of fluorescence lifetime changes of the retina before and after cataract surgery. Additionally, fluorescence lifetimes of the lens were investigated and correlated with the severity of cataract.

The Influence of Lens Opacities

Lens opacities predominantly influence the fluorescence lifetimes of the SSC (498–560 nm). This was less obvious in the LSC (560–720 nm). Emitted fluorophores with shorter wavelengths might be more absorbed or scattered by lens opacities than those with longer wavelengths, leading to a prolongation of lifetimes in the SSC. Furthermore, the transmission of light through the lens notably decreases with age, especially for the lower wavelengths.¹⁹

The Influence of the Excitation Laser

The excitation laser with a wavelength of 473 nm may be influenced by the reduced spectral transmission of shorter wavelengths in elderly patients.¹⁹ However, possible absorption or scattering of the excitation beam should result in the same proportion of changes

in both spectral channels and cannot explain for the difference between the two detection channels.

Regional Differences in Lifetimes

Lens opacities predominantly influenced the fluorescence lifetimes within the macular center. In a healthy eye, the macula features the shortest lifetime values compared with the surrounding retina. However, in subjects with cataract, these foveal lifetimes can be longer compared with those of the surrounding retina (Table 2). In eyes with cataract, increased scattering of light and additional contribution of lens fluorescence may lead to a significant prolongation of the recorded fluorescence lifetime from the macular center. Investigating the corresponding MPOD measurements a similar significant difference before and after cataract surgery can be found at 0.5° and at 2° eccentricity.

Influence of Various Types of Lens Opacities

The scores NO and NC of the LOCS III cataract grading scheme correlated significantly with the observed retinal *Tm* changes. The C score did not show a significant correlation with the retinal fluorescence lifetime difference. A possible explanation for this would be that cortical opacities usually commence at the equator of the lens and do not obstruct the center. As the ETDRS grid represents the macula only up to a radius of 3 mm from the fovea, cortical opacities might not interfere with photons passing through the center or might solely induce a mild scatter effect.

The P score showed a significant, although weak, correlation. Subcapsular posterior cataract in our cohort was usually present in the center region, and can be very dense, leading to a distinct drop in visual acuity and thereby also in the amount of photons passing. The rather high P value of patients with P in our study is probably influenced by the relatively small sample number (the LOCS III scores of all 39 graded lenses can be found in Supplementary Data 2).

Calculation of a Correction Factor Using LOCS III

In summary, the linear correlation of longer fluorescence lifetimes with increasing NO score can provide the following conversion equation:

> $Y_{SSC(Total)} = 197.8 * NO - 314.7$ $Y_{LSC(Total)} = 55.9 * NO - 93.65$

where Y represents the expected change in mean retinal fluorescence lifetimes (*Tm*) in ps (in the SSC or LSC and for the total area of the ETDRS Grid), and NO stands for the nuclear opacity score of the LOCS III cataract grading scheme. The correlation graph for both equations is drawn in Figure 5.

The X-intercept being at 1.6 and 1.67 for the SSC and LSC, respectively, leads to the conclusion that a lens with an NO score of 1.6 or less does not lead to significant changes in retinal fluorescence lifetimes, and is grossly indifferent to an artificial lens regarding FLIO measurements. This means in phakic eyes with an LOCS III NO score of 1.6 or less, no correction of fluorescence lifetimes is necessary.

In the foveal region a stronger decline in lifetimes after surgery was observed. The equation above accounts for the total area of the ETDRS grid. Therefore when utilizing this formula, lifetimes in the fovea are probably undercorrected, and lifetimes in the outer ring might be overcorrected. If certain areas of the retina are of interest, a specific conversion formula for each grid area (center, inner ring, outer ring) should therefore be used (see Supplementary Data 1).

Crystalline Lens Fluorescence Lifetimes

FLIO measurements of the crystalline lens featured significantly longer Tm compared with measurements of the artificial lens in both spectral channels. Furthermore, more opaque crystalline lenses exhibited comparatively shorter *Tm* when compared with clearer crystalline lenses (using NO score as measure). One hypothesis might be that lens fluorophores normally feature long lifetimes. With advancing age and lens opacity, the lifetime of the lens fluorophores decreases, however, the lens increasingly contributes to light scattering. We tested this theory by correlating the NO score against the respective lens Tm: effectively higher NO scores correlated significantly with lower lens Tm (P < 0.0001 for both spectral channels, $r^2 = 0.4$ and 0.52 for SSC and LSC respectively; see Fig. 6, bottom row).

Thereby the significant correlation between the crystalline lens Tm and the change in retinal Tm after surgery is well explained (Fig. 6, top row). In addition, this correlation enables the calculation of similar conversion formulas as the ones based on LOCS III grading as an alternative approach.

Other Approaches on Correcting for Lens Opacities

There have been various other approaches to solve the issue of artifacts induced by the crystalline

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Figure 6. Correlation of the mean fluorescence lifetime of the crystalline lens (lens *Tm*) with the mean change in retinal lifetimes (retinal *Tm*) after cataract surgery in the upper row, and with the NO subscore of the LOCS III cataract grading scheme in the bottom row (SSC on the left, and LSC on the right). The correlations illustrate the negative association of lens *Tm* with cataract, meaning longer lens *Tm* represent clearer, less opaque crystalline lenses, and shorter lens *Tm* are associated with higher NO scores.

lens fluorescence on retinal lifetimes. Klemm et al.²⁰ proposed using separate measurements of the crystalline lens to measure its contribution to the fluorescence lifetimes. However, grading of the crystalline lens provided, based on our data, a more reliable correction.

Another approach, applied directly during image acquisition, is through an optimized optical arrangement based on the proposition of Schweitzer et al.^{21,22} With an annular ring, a reduction to 4% for the lens fluorescence can be achieved while reducing the simulated retinal fluorescence to 42%. These data are based on a model eye simulating retinal and crystalline lens fluorescence. A disadvantage of this approach is the roughly doubled acquisition time because of the attenuated fluorescence signal detection, although there is no separate lens measurement or grading of the lens necessary. This solution would be less prone to bias than our method, although its practicability in vivo has yet to be verified. Only recently, Brauer et al.²³ also examined fluorescence lifetimes of 31 patients before and after cataract surgery and compared the resulting differences in lifetime with four predictive computational models, whereby a three-exponential model with a time-shifted lens fluorescence resulted in the best fit.

Limitations

Limitations of our study are the low patient count. In two patients both eyes were included. Although only the intraeye difference was examined, which largely counteracts this shortcoming. Furthermore, the interval for the postoperative measurements was rather short with 1 week. We verified the absence of relevant media opacity and significant inflammation, and all surgeries were performed by highly experienced surgeons leading to very low postoperative inflammations. Although slight amounts of subclinical postoperative media opacification and/or inflammation could have an influence on the results. Seven different models of lens implants were used, which could have an effect on the outcome. Unfortunately, we did not have enough eyes in our study to create a meaningful comparison between the individual implant types and changes in lifetimes before and after surgery.

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Conclusions

Lens opacities result in a significant prolongation of measured retinal fluorescence lifetimes in both spectral channels. We found a significant linear correlation of the retinal fluorescence lifetime with the grade of the nuclear opacification and the fluorescence lifetime of the opaque lens and propose a conversion formula to correct for FLIO measurements in eyes with cataract.

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