Field-of-view Experiment summary - FISH shadowing experiment - Sep 2012

Cruise: Oahu-13
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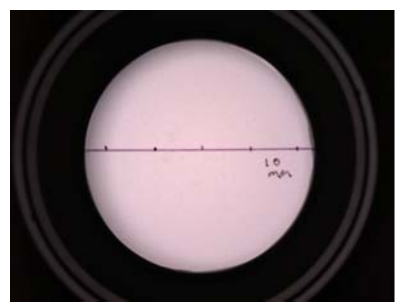
January 8, 2013

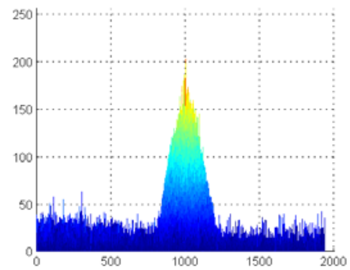
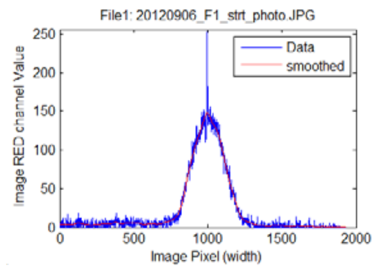
This document will detail all the Field-of-view measurements done on the FISH used during the 2012 Sept shadowing experiment. First I will describe each of the 3 experiments then I will show data and discuss results. Three types of measurements were made 1) images of the light from the fiber 2) scanning a changing-diameter iris aperture and 3) scanning the OL455 exit port aperture whilst changing the FO-to-aperture distance.

<u>Fiber tip light images</u>

In this experiment Mark built a jig which allows a fiber to be attached at a fixed distance from a diffuser. The distance from the fiber tip to the diffuser screen is 28.5 mm. Diffuser screen material is fine grit (AR5D) silica carbide fiber optic polishing film 0.005" thick with the abrasive side facing the fiber input. Once the fiber was attached light was shown in the open end of the fiber and illuminated the diffuser. Mark then used his iPhone to take a photo of the diffuser from a fixed distance. Centering of the fiber and diffuser reference scale images were done by eye, so indexing is imperfect between images.

		
FOV jig: The fiber is attached at the bottom. The diffuser is in the middle (not shown) and the iPhone is positioned at the top.	Photo of the fig from the iPhone end.	Image used to scale the fiber images from pixels to millimeters. The scale shown is marked off at 10 mm increments. This picture is taken at the same camera to diffuser distance as the fiber images.

Once the jig was ready Mark and Carol took photos of each fiber. And produced 7 jpgs images, one for each fiber.

		
Fiber number 1 image taken on 6 Sept 2012	Side view of the fiber 1 image with the red image (green and blue images not shown).	Red image in cross-section with the ssa smoothed data.

The JPG images for each of the 7 fibers were read into Matlab as red,green and blue mapped images and analyzed. In Matlab the JPG is a 1936 by 2592 by 3 array containing the red, green and blue image data. The analysis of the 7 photos was done a few different ways.

1) **Single red image**: In this initial step just the red part of the image was used. And one row across the image was pulled out and smoothed (right most figure above). The row was chosen at the center of the peak.   I then found the width of the peak at the base of the peak. One number for each of the photos. The red, green and blue images for each photo looked fairly similar so for this intial analysis the red image was used.

2) **Summed images at 15 degree angles**: The next attempt was to do the same as in step 1 but with a sum of the red, green and blue images and the slice across the image would be in 15 degree increments. So you would take a slice to get the peak and then rotate the image 15 degrees and repeat until the image had been rotated 360 degrees. This would give you 25 estimates of the peak width for each of the 7 photos.

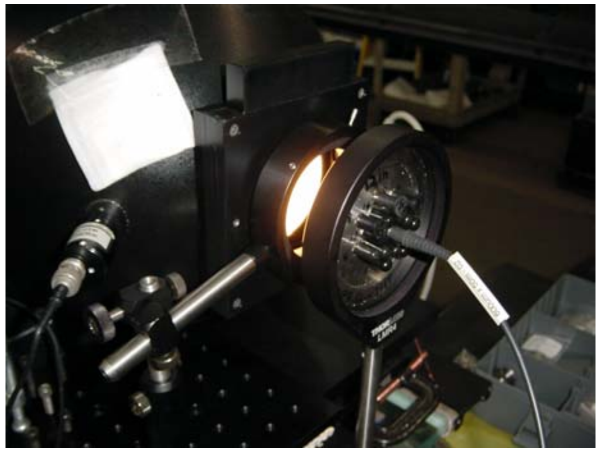
3) **Fit a circle**: In this iteration I used a contour plot to pull out the data for the lowest usable contour. Then I fit this data to a circle. This was done for the red, green and blue images in the photo. So for each photo you get 3 estimated widths (one estimate for the red, green and blue images).

Methods 1 and 2 could underestimate the width because it could miss the real lowest value. The circle should kind of average out the shape differences around the peak base. But could also underestimate the width for the same reason at methods 1 and 2. If the contour is not at the lowest value then the circle will be fit too high and underestimate the width.

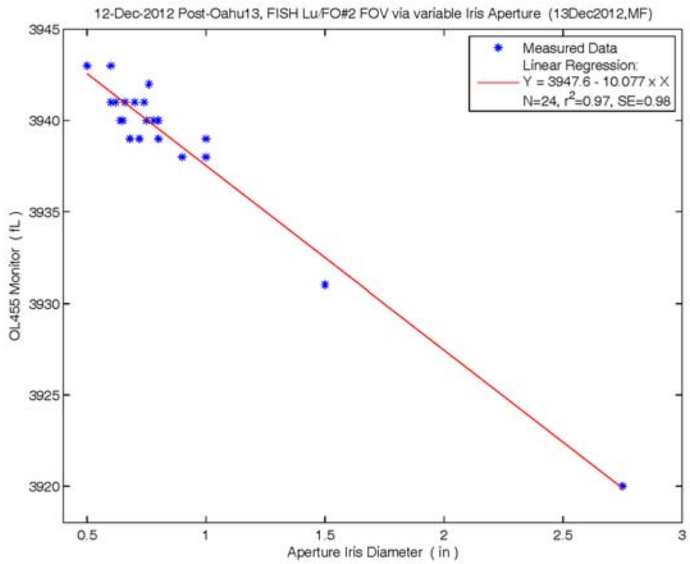
I should mention that in all these methods finding the center of the image was problematic. You could not use the max value because of large spikes at the top of the peak. This would affect method 2 the most. Because trying to aline the 25 different images to average them was difficult.

Scanning a changing-diameter iris aperture

In this experiment Mike took the OL455 sphere and mounted mounted a variable-diameter iris and the fiber optic jig in front of the sphere aperture. Measurements where taken with the FISH as the diameter of the iris aperture was changed. With a fixed distance between the fiber and the iris opening the math is pretty simple to figure out the angle.

	
Setup with OL455 sphere and fiber optic jig	Fiber optic jig mounted in front of the OL455 with one fiber in the middle of the jig

During the experiment data were collected on the OL455 sphere monitor. Mike noticed that as the iris closed the sphere got brighter. A regression of the monitor data and the distance allows us to correct the lamp values for this increase.



All the FISH data were adjusted using the regression in this graph. The sphere monitor change min-to-max was 0.59 %.

I should note for this experiment and the following experiment there is blue spectrograph and red spectrograph data. The two specs are using the same fiber optic cable (See figure 10).

Also this was done on fiber optic # 2 which is Track 3.

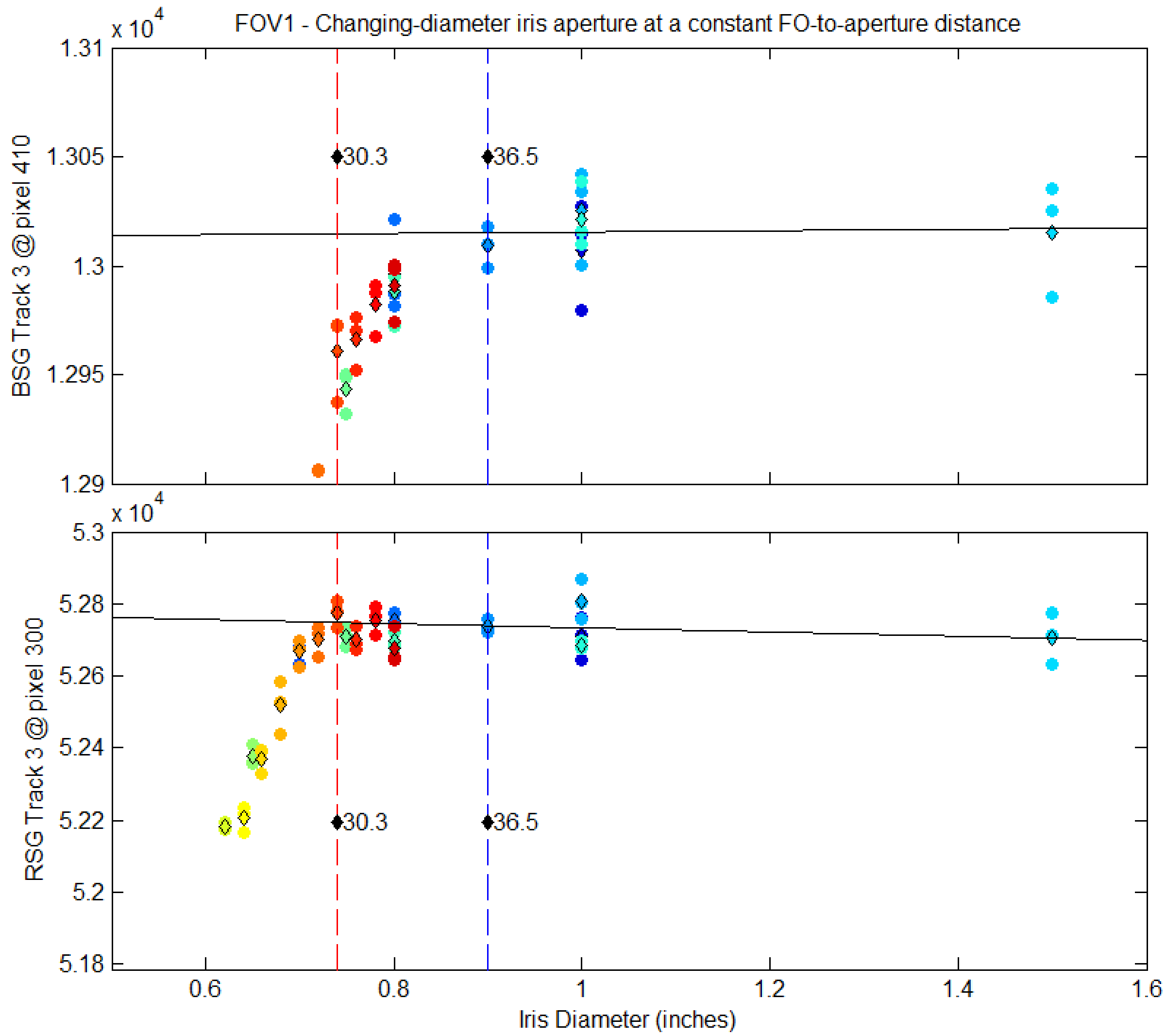
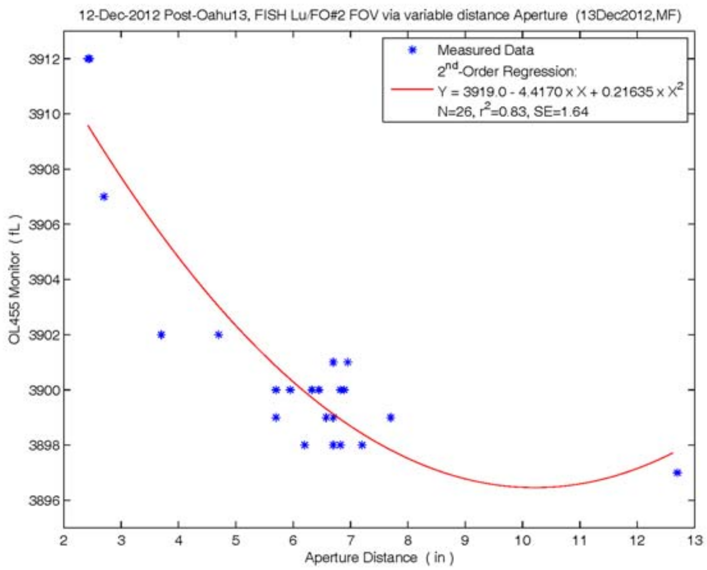


Figure 10. Plot of FISH blue and red spectrograph data for the changing-diameter aperture experiment. For the blue spectrograph pixel 410 (586 nm) at the data peak and for the red spectrograph pixel 300 (792 nm) was used.

The above graph shows the data from this experiment. I picked the pixel at the peak for each spectrograph and plotted them verse Iris Diameter. The idea is that as the iris is closed the amount of light getting into the FISH through the fiber will decrease when the iris is no longer completely in the FOV of the fiber. I then used the data to find the iris diameter right before any decrease in signal and use this value to calculate the FOV. In the graphs above I chose the last data point before I saw any decrease. Interestingly this was different for the blue and red spectrograph.

Scanning the OL455 exit port aperture whilst changing the FO-to-aperture distance

In this experiment the setup is similar to the above setup except the iris was removed and the and the sphere exit port distance from the fiber optic jig was varied.



All the FISH data were adjusted using the regression in this graph. The sphere monitor change min-to-max was 0.39 %. ?

I should note for this experiment and the following experiment there is blue spectrograph and red spectrograph data. The two specs are using the same fiber optic.

This experiment was preformed twice. Once on 12 Dec on Fiber Optic cable #2 (Track 3) and again on Dec 14th on Fiber Optic cable #7 (Track 13). Dec 14th sphere monitor change min-to-max was 0.41 %.

The following graph (Figure 12) shows the data from the 12 Dec experiment. I picked the pixel at the peak for each spectrograph and plotted them verse FO-to-aperture distance. The idea is that as the distance from fiber to aperture increases the amount of light getting into the FISH through the fiber will decrease when the aperture is no longer completely in the FOV of the fiber. I then used the data to find the distance right before any decrease in signal and use this value to calculate the FOV. For the red spectrograph I chose the last data point before I saw any decrease. In the case of the blue spectrograph I had to guess were there the data would start decreasing. Interestingly this was different for the blue and red spectrograph.

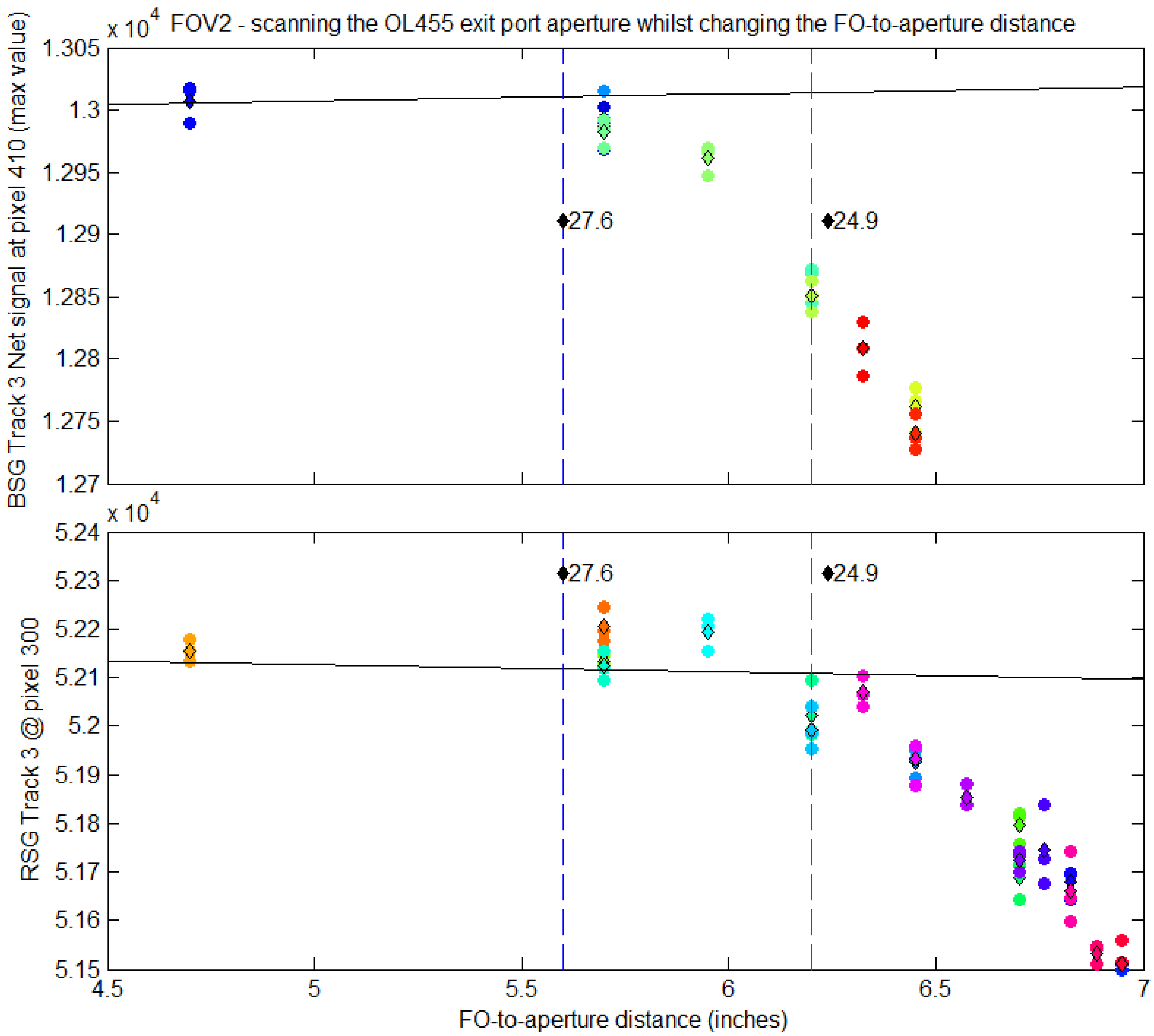


Figure 12. Plot of FISH blue and red spectrograph data for the changing FO-to-aperture distance experiment - 12 Dec. For the blue spectrograph pixel 410 (586 nm) at the data peak and for the red spectrograph pixel 300 (792 nm) was used.

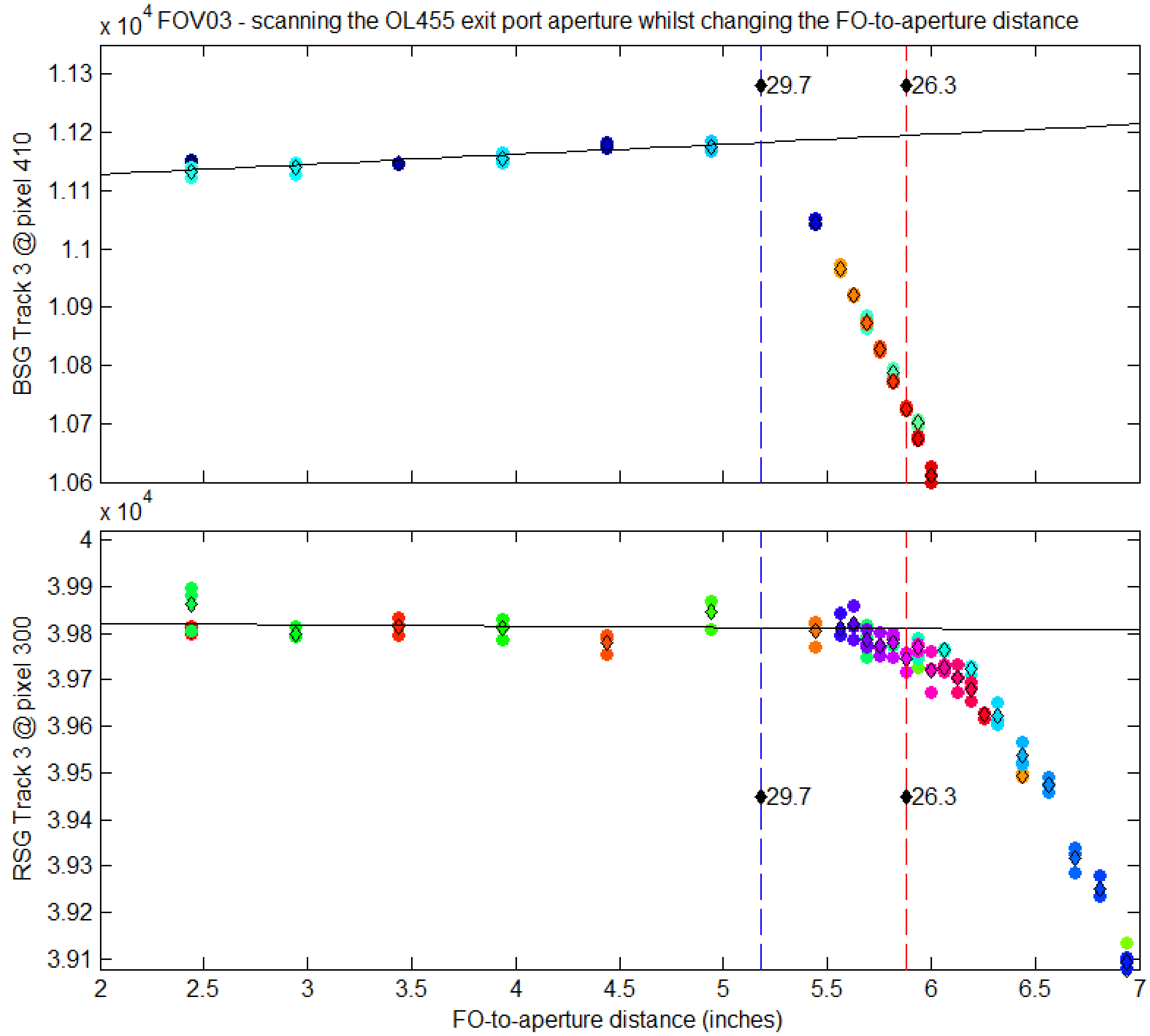


Figure 13. Plot of FISH blue and red spectrograph data for the changing FO-to-aperture distance experiment - 14 Dec. For the blue spectrograph pixel 410 (586 nm) at the data peak and for the red spectrograph pixel 300 (792 nm) was used.

The above graph (Figure 13) shows the data from the 14 Dec experiment. I picked the pixel at the peak for each spectrograph and plotted them verse FO-to-aperture distance. The idea is that as the distance from fiber to aperture increases the amount of light getting into the FISH through the fiber will decrease when the aperture is no longer completely in the FOV of the fiber. I then used the data to find the distance right before any decrease in signal and use this value to calculate the FOV. For the red spectrograph I chose the last data point before I saw any significant decrease. In the case of the blue spectrograph I had to guess were there the data would start decreasing. Interestingly this was different for the blue and red spectrograph.

Results

All FOV angles presented below are full-width angles.

Fiber tip light images:

Single red image

Fiber#	**FOV (Degrees)**
1	25.52
2	23.87
3	26.64
4	29.27
5	28.04
6	27.40
7	28.22

http://data.moby.mlml.calstate.edu/moby2_testing/20120903_shadowing/resonon/fiber_images/plt_20120906_Fx_str_imgs_2mm__/plt_20120906_Fx_str_imgs_2mm__.html

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Summed images at 15 degree angles:

Fiber#	**Mean (N=25) FOV (Degrees)**	std	%std
1	24.94	0.43	1.72
2	23.04	0.31	1.33
3	26.28	0.49	1.85
4	28.62	0.69	2.41
5	26.04	0.42	1.62
6	27.23	0.48	1.78
7	26.39	0.31	1.17

http://data.moby.mlml.calstate.edu/moby2_testing/20120903_shadowing/resonon/fiber_images/plt_20120906_Fx_str_imgs_2mm_slice_/plt_20120906_Fx_str_imgs_2mm_slice_.html

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Fit a circle

Fiber#	**Mean (N=3) FOV (Degrees)**	std	%std
1	31.27	0.37	1.20
2	31.93	0.06	0.19
3	31.15	0.32	1.02
4	30.79	0.11	0.37
5	31.55	0.27	0.87
6	31.31	0.79	2.53
7	30.99	0.52	1.68

http://data.moby.mlml.calstate.edu/moby2_testing/20120903_shadowing/resonon/fiber_images/p lt_20120906_Fx_str_imgs_2mm_contour3_fit_/plt_20120906_Fx_str_imgs_2mm_contour3_fit_.html

<u>Scanning a changing-diameter iris aperture</u>

	BLUE	RED	
Dec 12 exp 1	36.5	30.33	iris changing (FO#2)

<u>Scanning the OL455 exit port aperture whilst changing the FO-to-aperture distance</u>

	BLUE	RED	
Dec 12 exp 2	27.59	25.00	FO-to-aperature distance (FO#2)
Dec 14 exp 3	29.7	26.32	FO-to-aperature distance (FO#7)

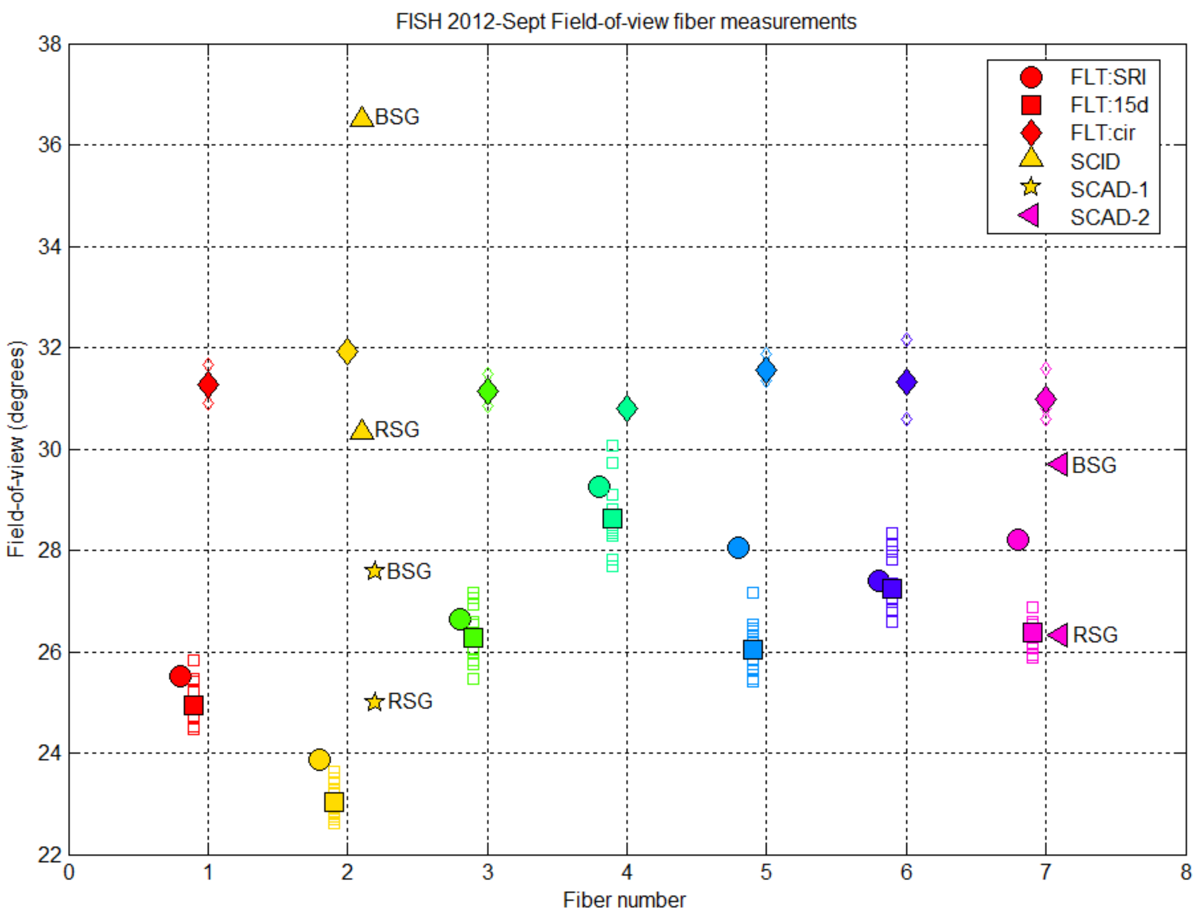

Figure 14. Figure showing all the estimates of FOV for each fiber and each experiment.

All the Experiment FOV estimates. Large symbols with the black outline are the mean of that experiment. Each symbol is an experiment. Colors are the fiber number (symbols for each fiber are offset so the symbols don't overlap). Smaller open symbols are the individual numbers used to calculate the mean (if they exist for the experiment).

FLP:SRI = Fiber tip light images: Single red image, FLP:15d = Summed images at 15 degree angles, FLP:cir = Fit a circle, SCID = Scanning a changing-diameter iris aperture, SCAD-1 = Scanning the OL455 exit port aperture whilst changing the FO-to-aperture distance (FO#2) and SCAD-2 = Scanning the OL455 exit port aperture whilst changing the FO-to-aperture distance (FO#7).

Not sure what conclusions to make about the FOV. Each experiment gave different answers. The iris diameter experiment seems to be the largest outlier and the fitted circle seems to be the next. The fitted circle also gave me most consistent answers for all fibers.. FO#2 had the most experiments and vary the most as well. Not sure what else to conclude.