RECORDING FAST OPTICAL DATA

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Prior to Data Collection…

Design an Experiment

Determine what brain regions you need to sample

Design a montage (placement of sources and detectors)

Check source/detector distances for cross-talk

Implement the experiment with software that can send out a trigger and/or marker to synchronize stimuli with Boxy recording

Set Boxy to record continuously for the length of 1 block (and set all other Boxy parameters)

Check the timing consistency between behavioral expt and Boxy recording

Time Division Multiplexing of Sources (aka Muxing)

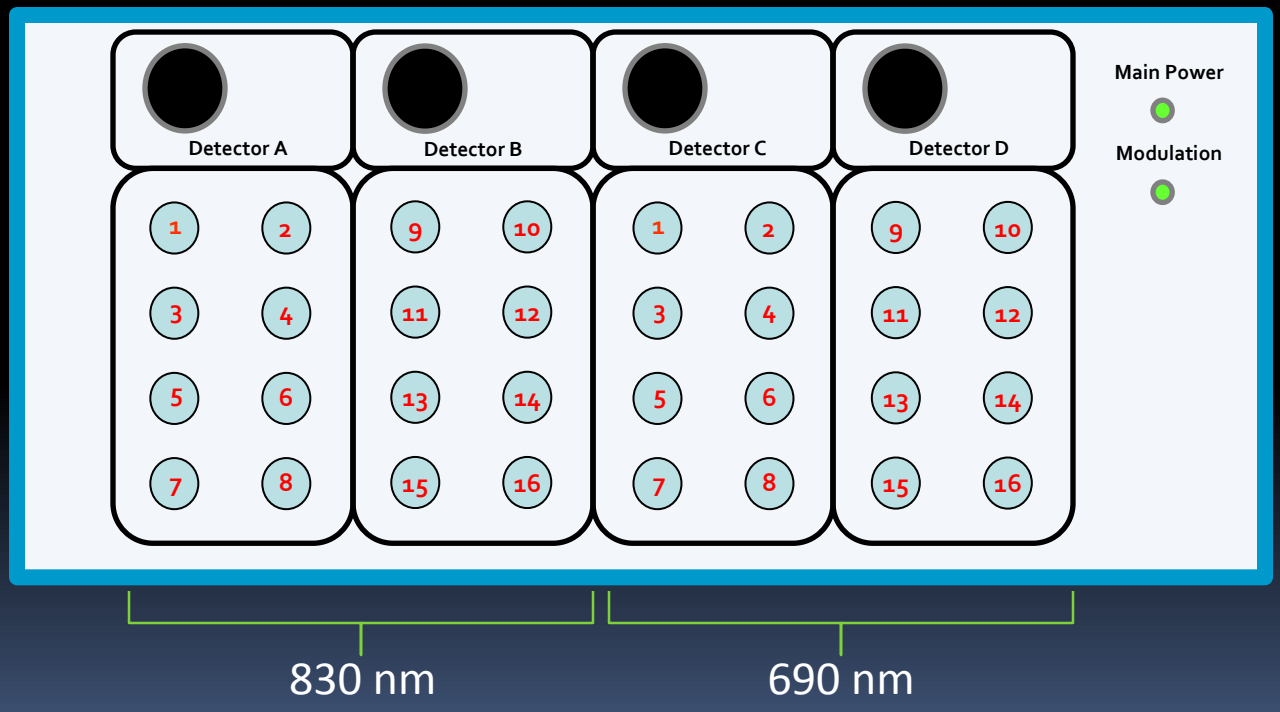
- Not all sources are on at the same time, instead they come on in series
- We call each slice of time that a source is on a “mux channel” or “mux slot”
- The PMT (detector) cannot differentiate sources, it will collect all light available during any given time
- Therefore, you must take care when placing sources that share a mux slot (i.e., 2 sources that turn on at the same time)

Switch Settings on Imagent

Switch 16 mode

Maximum of 2 sources on at a given time (per Imagent)

Maximum of 16 MUX channels

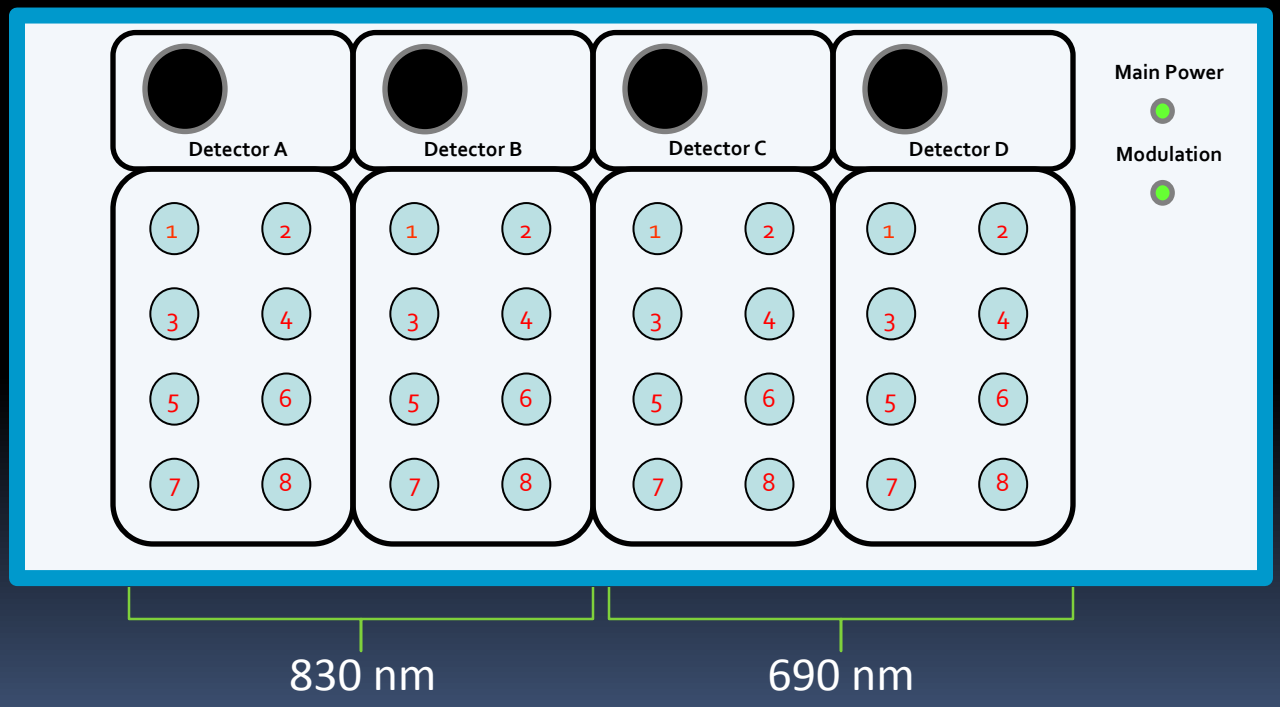


Switch Settings on Imagent

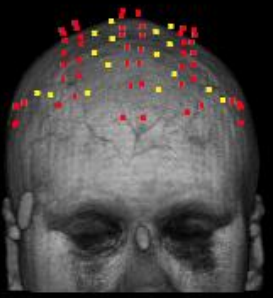
Switch 8 mode

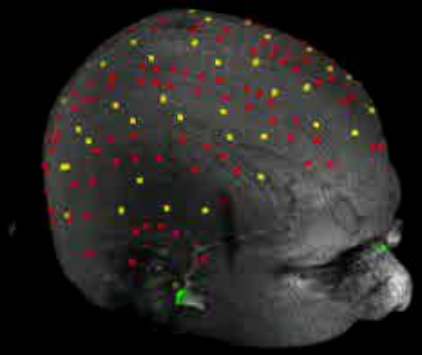
Maximum of 4 sources on at a given time (per Imagent)

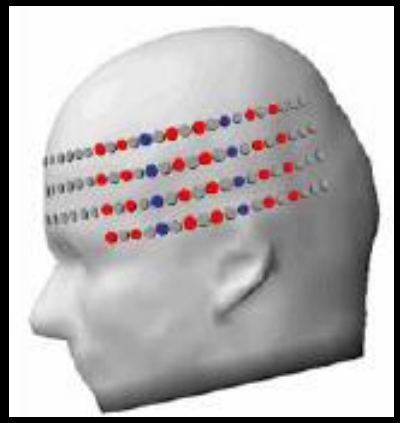
Maximum of 8 MUX channels

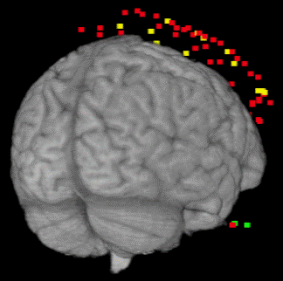


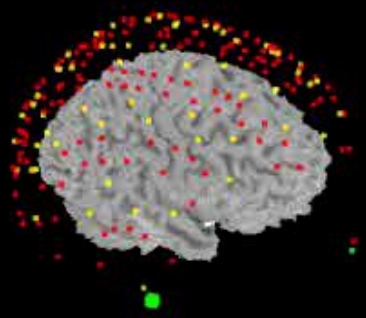
EROS Montages

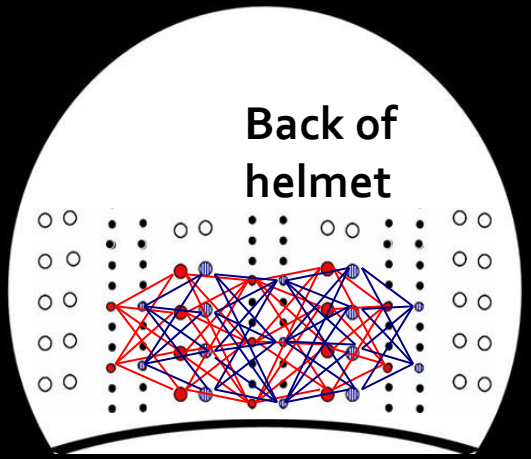










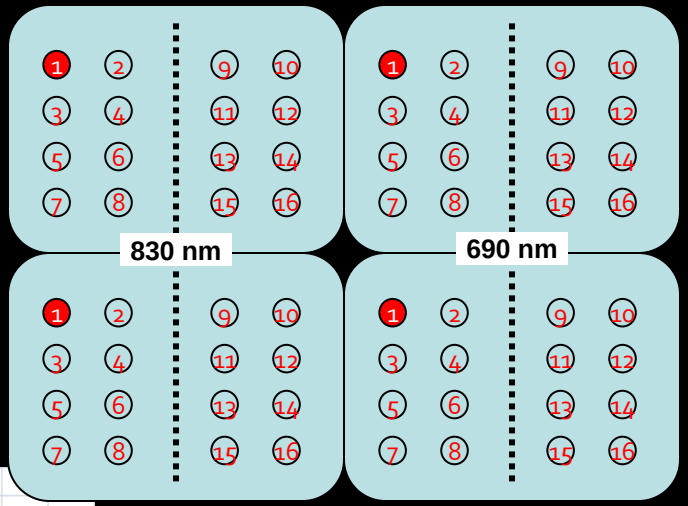


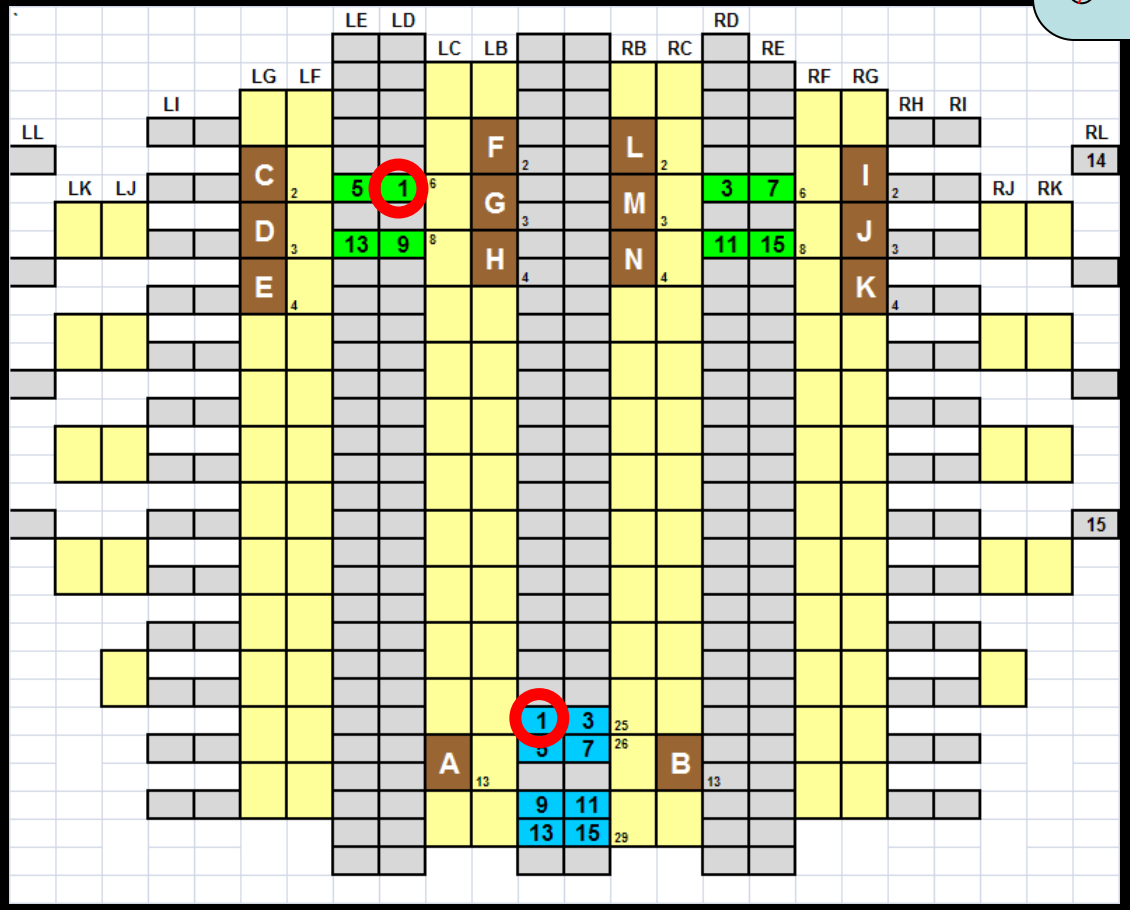
Considerations for Montage Design

- The more crossing/overlap of channels, the better the S/N and the better the localization.
- The more varied the src-det distances, the better coverage across cortical depths.
- **To avoid cross-talk**: when 2 or more diodes are turning on during the same MUX slot, only 1 can be within 6 cm of a given detector
  - This distance will be smaller with certain populations (older adults, infants) due to increased transparency.
- Optimize design such that most src-det pairs have a distance of 2-5 cm (most at 2.5 - 3 cm)
  - Too short, not hitting cortex
  - Too long, lose too many photons

Example:

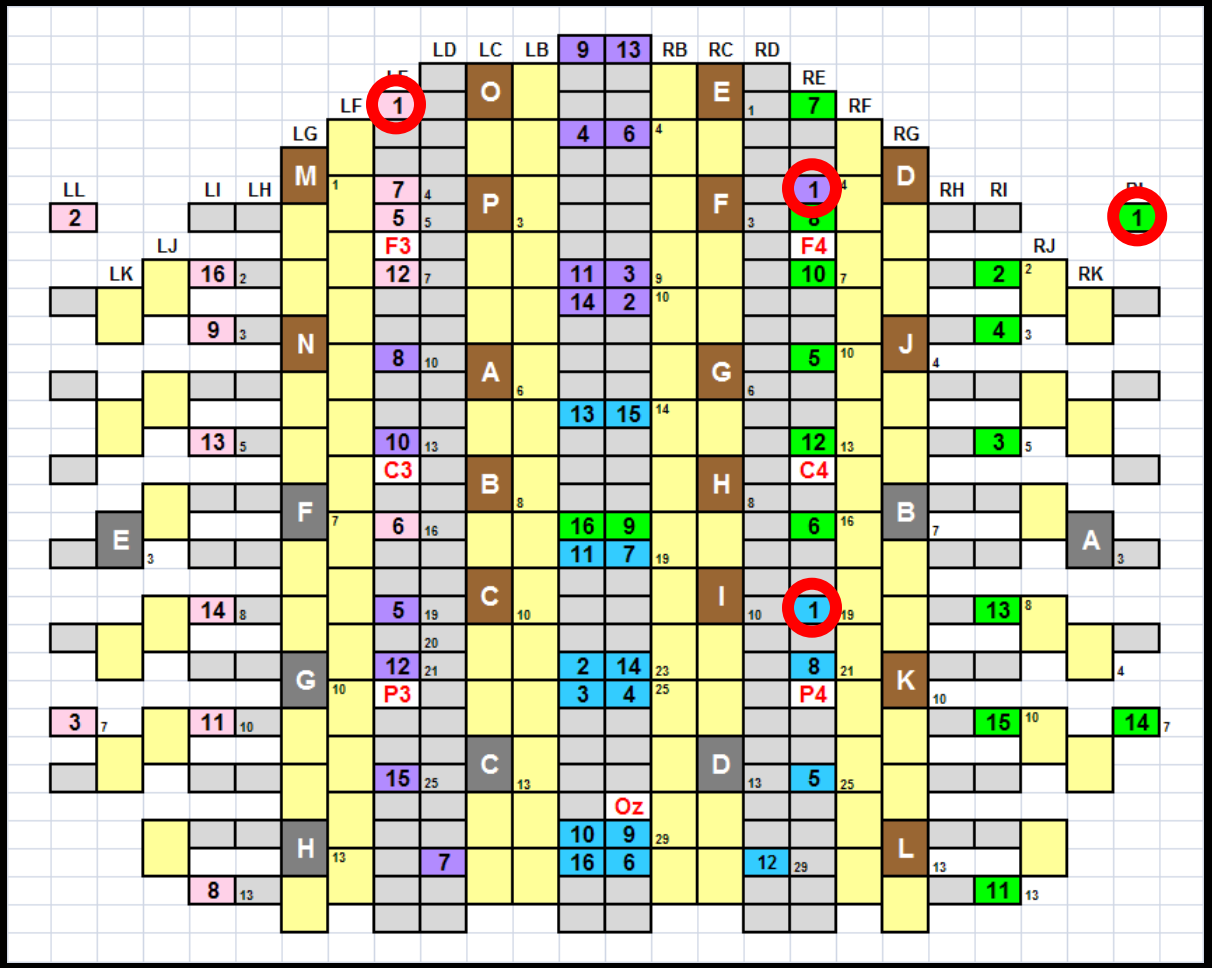
Dual wavelengths (2 sources in each hole)
Three patches (bilateral frontal and occipital)

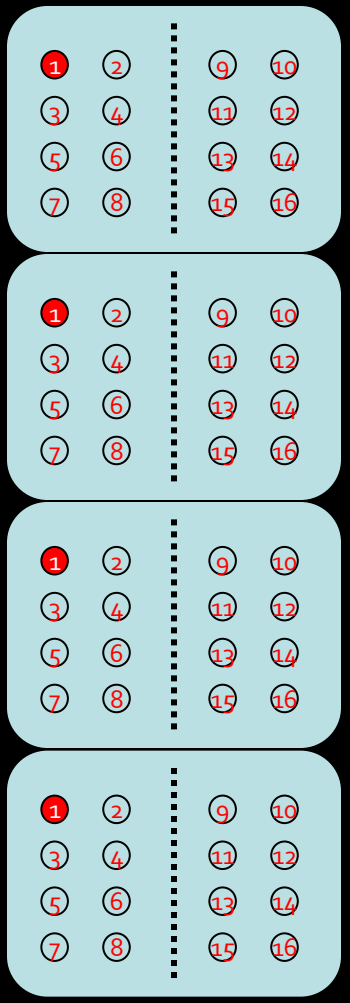




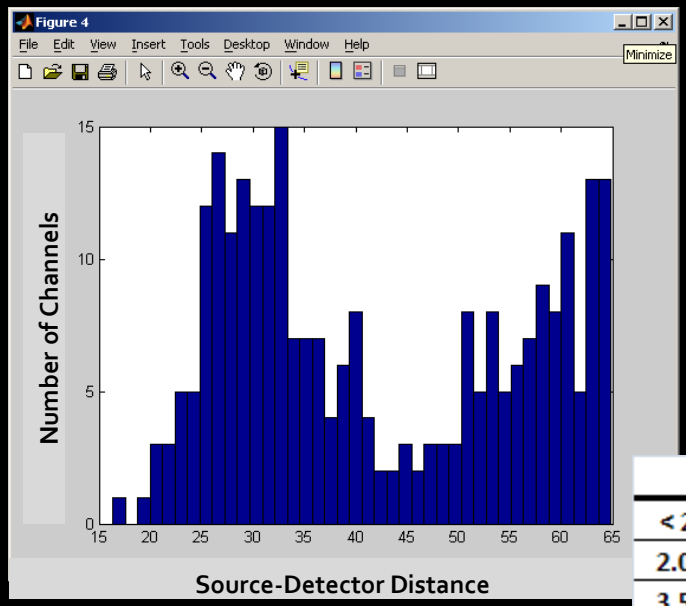
Example:

- One wavelength (1 source in each hole)
- Almost full cortical coverage
  - 24 Detectors
  - 60 Sources (mux 16)





All 830 nm

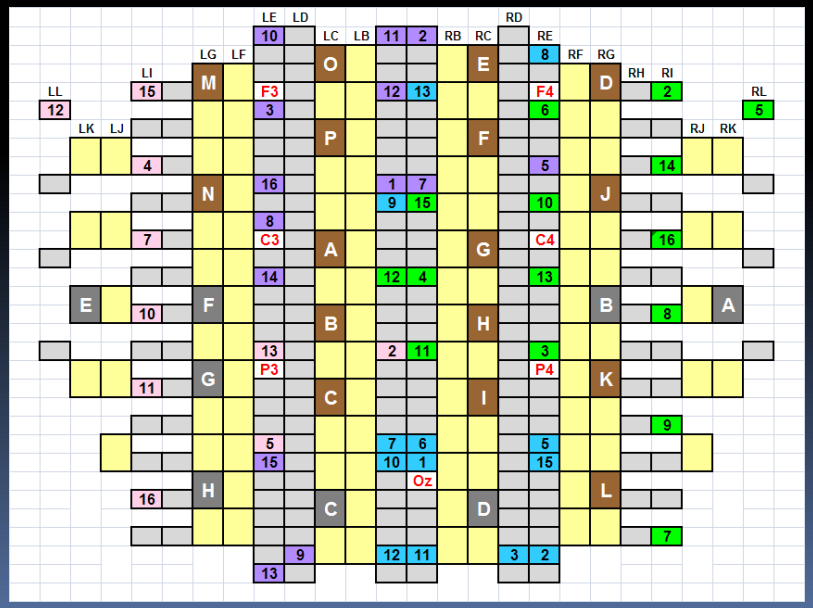


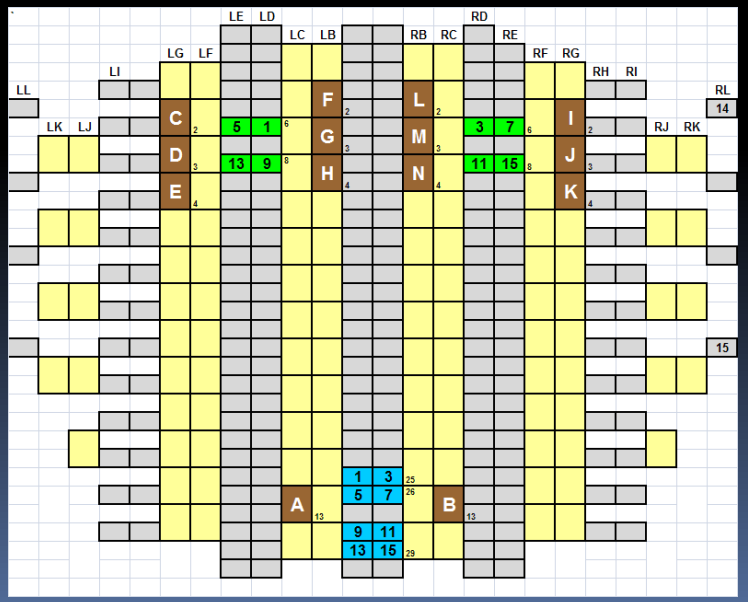
Full Montage

	Number	%
< 2 cm	1	0.3%
2.0-3.5	118	30.7%
3.5-4.5	38	9.9%
4.5-5.5	39	10.2%
>5.5	188	49.0%
total	384	

Patches

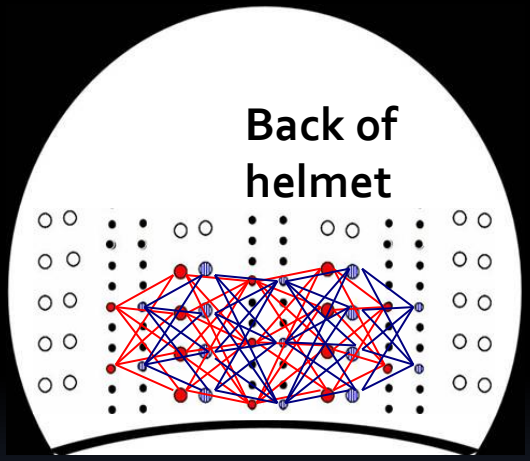
	Number	%
< 2 cm	2	0.9%
2.0-3.5	102	45.5%
3.5-4.5	38	17.0%
4.5-5.5	26	11.6%
>5.5	56	25.0%
total	224	





Why do we use layouts based on rows?

- Maximizes the spatial sampling across depths
- Maximizes the overlap of channels

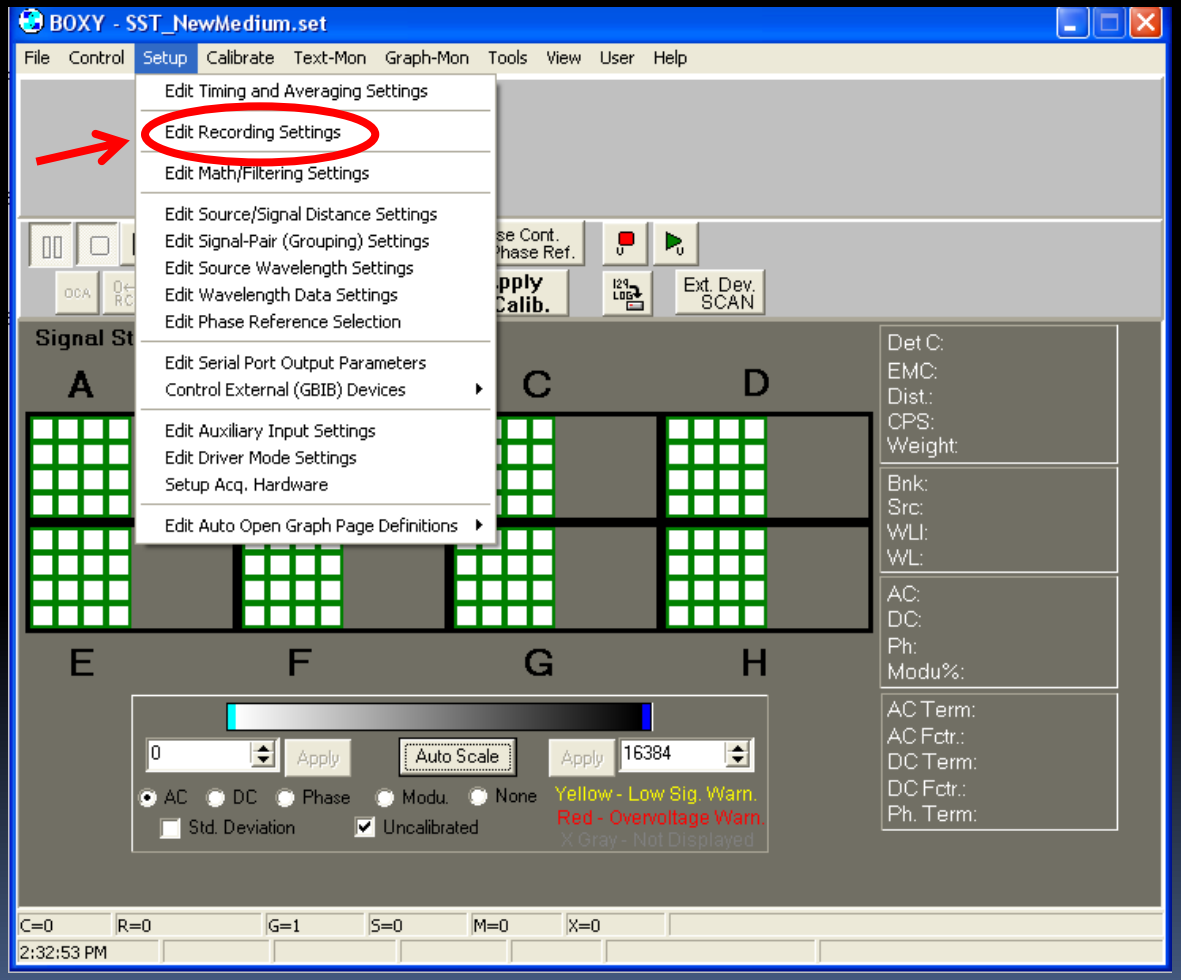


- For large montages, we have scripts to help automate the process of assigning mux channels while avoiding cross-talk (see Kyle Mathewson and Ed Maclin)
- Under development: using multi-objective optimization theory to design montage configurations automatically using a genetic algorithm to search the space of possible montage configurations and return solutions achieving localized sampling and cross-talk minimization objectives (see David Steines and Pauline Baniqued)

Syncing stimulus presentation with data collection

- Boxy receives a trigger to start recording data
  - Via BNC connection
- Boxy can also record a digital code that can mark the timing and type of specific events in the data stream
  - Via a 9 pin D-connector
  - These codes appear in the data file under the column labeled digital auxiliary (on mux 1 only)
  - We cross check this timing with the output of our experiment presentation software

BOXY: Imagent data collection software



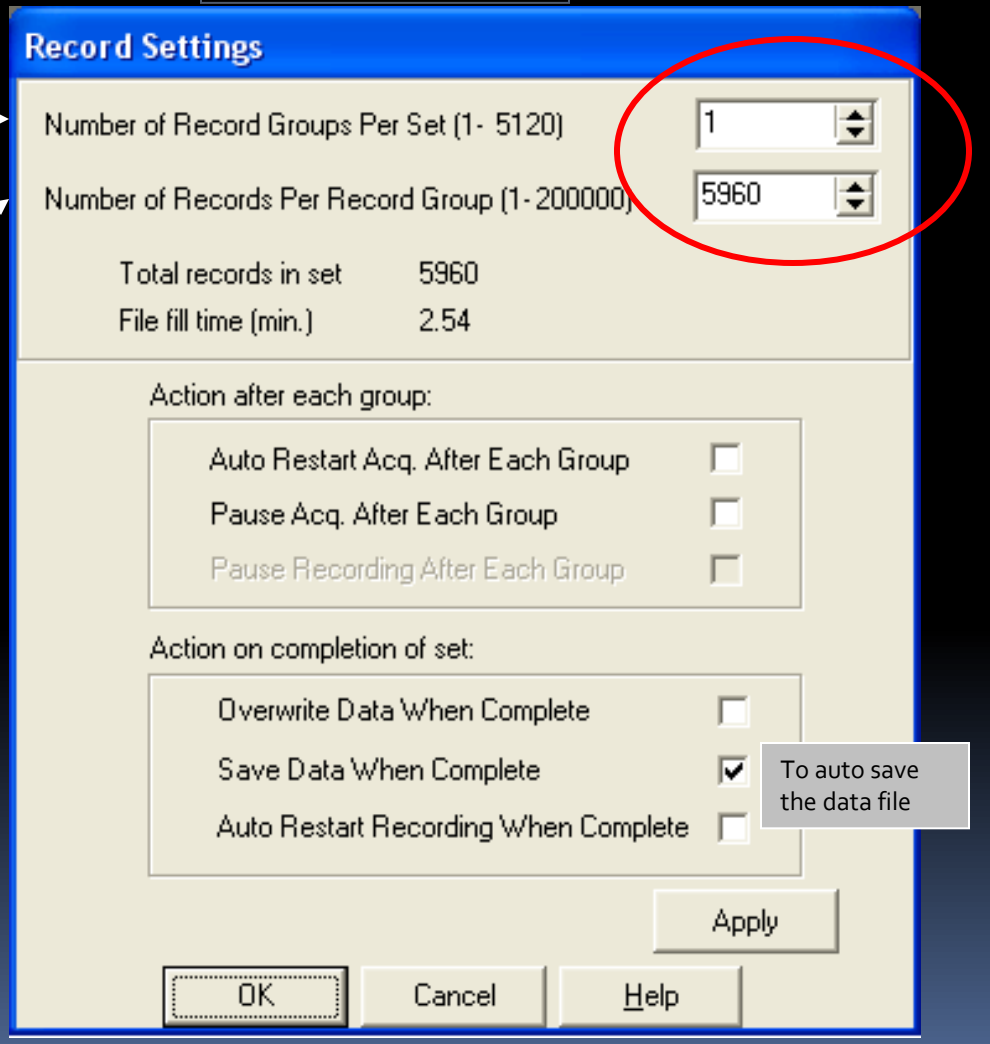
Setting the Recording Window in Boxy (Edit Record Settings)

Record = data point or sampling point

This will usually be set to 1 because we will re-trigger Boxy with every block .

This will be determined by the timing of a single block.

"File fill time" should be a little longer than the time it take to complete 1 experimental block.



To auto save the data file

Ex. Optical sampling rate = 62.5 Hz ; Block duration = 3.3 min

Sampling period = 1000/62.5 = 16 ms

12600 pts * 16 = 201,600 ms = 201.6 sec = 3.36 min

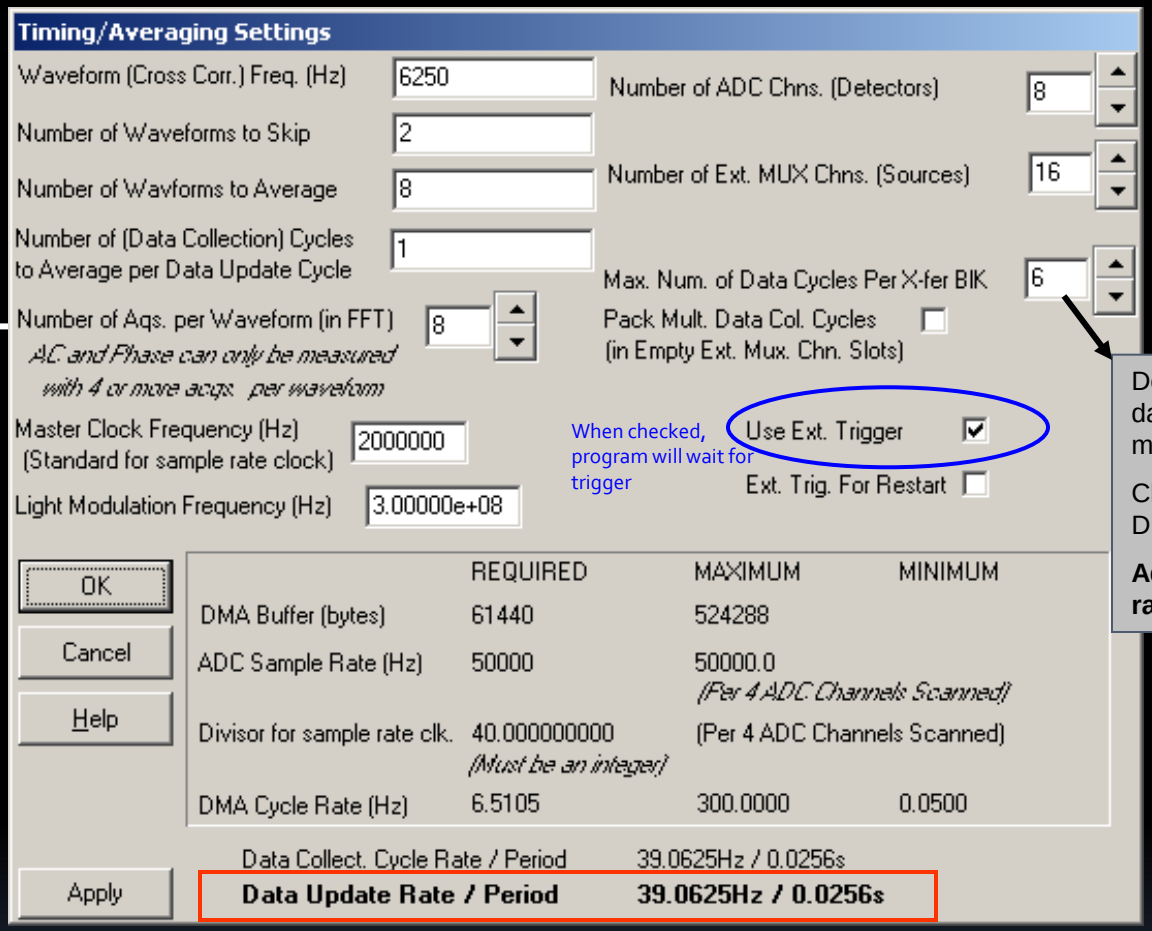
→optical will collect data continuously for 3.36 minutes

Timing/Averaging Settings in Boxy

"Waveform" refers to 1 cycle of the CCF

Number of samples/cycle of the CCF

Will change ADC Sample Rate, Divisor, and DMA rates



Determines how often the data are transferred to memory (X-fer = Transfer)

Changes DMA Buffer and DMA cycle rate

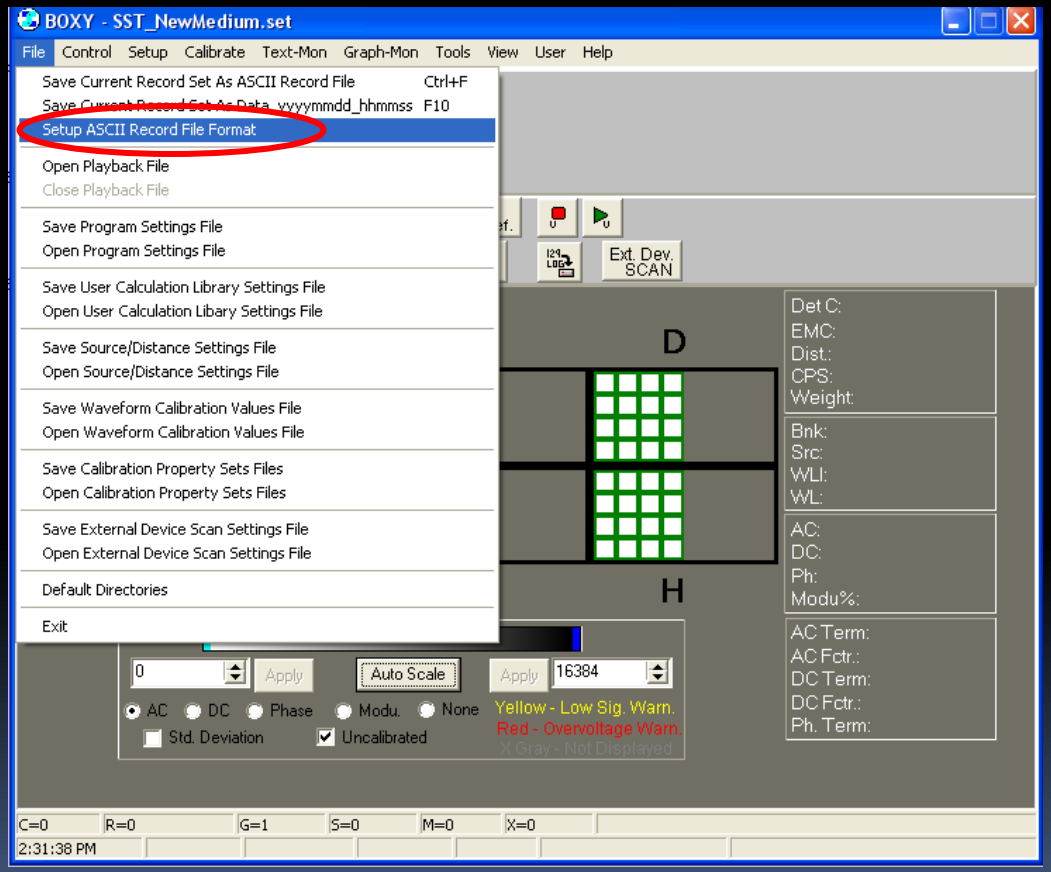
Adjust so that DMA cyle rate is b/w 5-10 Hz

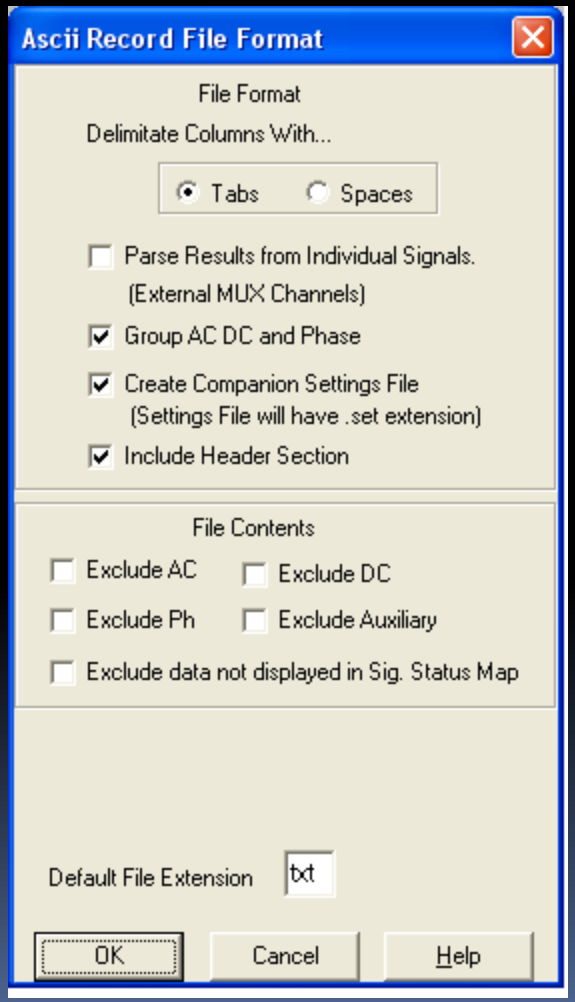
Data Update Rate = final sampling rate

The final sampling rate is determined by:

1. **The number of sources (mux channels)**
2. **The cross-correlation frequency**
3. **The number of waveforms to skip**
4. **The number of waveforms to average**
5. **The number of cycles to average per data update cycle**

File format settings in Boxy
--For compatibility with p_pod





Tips for recording that we've learned over the years…

- Support the fibers from above
  - Less weight on subject
  - Added support near the receiving end of the fiber
  - Less tangling/breakage of fibers
  - Adds clearance to the back of the head
- Don't keep graphs or text monitors open while recording data
  - Because it increases the risk of timing errors (i.e., missed packets)
- Limit the length of one block to less than 5 minutes, ideally less than 3
  - This greatly reduces movement artifacts because it give the subject a chance to squirm a little between blocks
- The subject monitor refresh rate CAN appear as an aliased frequency in the AC intensity (and produce artificially low phase variability)
  - Minimize by using black/dark backgrounds instead of white
  - Cover regions of helmet/patch that might allow stray light in
  - Inspect the frequency spectrum of your channels as a quality control measure
- The subject chair is an important piece of equipment
  - Comfortable!
  - No swivel, recline, or wheels

Checking signal quality just prior to data collection

- Adjust gain (bias) of each detector such that DC values are near 16000 (limited by the shortest distance channels)
  - This will minimize the risk of over-voltaging
    - The sum of AC, DC, and PH has to be less than the A-to-D converion range (32K)
- Good channels should have:
  - AC values above 100
  - PH standard deviations below 5
    - Your shortest channels should ideally have stdev less than 1.
    - The lower the stdev, the better your signal-to-noise
- If you don't have very many channels in your montage, you can also plot each channel as a time course (AC intensity) to check that a pulse is visible.
  - You may want to use some of Boxy's calculation features to filter the plotted data to isolate the pulse signal

Boxy output file - header

- Important header information (appears at the top of the file)

```
BOXY.EXE: ISS Oximeter R&D Program Version 0.40
Ascii Record File

#ACQ INFORMATION
8  Detector Channels
10  External MUX Channels
0  Auxiliary Channels
6250  Waveform (CCF) Frequency (Hz)
2  Waveforms Skipped
8  Waveforms Averaged
1  Cycles Averaged
8  Acquisitions per Waveform
62.5000  Updata Rate (Hz)

#FILE INFORMATION
FALSE -- External MUX Channel results are NOT parsed.
TRUE -- AC DC and Phase are GROUPED.
TRUE -- Companion Program Settings File CREATED.
FALSE -- AC data not excluded
FALSE -- DC data not excluded
FALSE -- Phase data not excluded
TRUE -- Auxillary Chn. data excluded
Tab delimiters
```

- After this info, there will be more header info, but this is for use when collecting absolute oxy/deoxy concentrations (i.e., multi-distance method with dual wavelengths)

Boxy output file - Data

#DATA BEGINS																				
record	exmux	A-AC	A-DC	A-Ph	B-AC	B-DC	B-Ph	C-AC	C-DC	C-Ph	D-AC	D-DC	D-Ph	time	group	step	mark	flag	aux-1	aux-2	digaux
1	1	2.817	15.22	180	119.3	264.8	180	1842	3165	205.8	1565	4440	85.45	10014.3	1	1	0	768	0	0	0
1	2	2.946	9.719	224.2	239.1	472.2	200.2	3020	5132	225.2	40.62	93.75	88.24								
1	3	2534	4644	166.5	4791	9098	126.5	9.868	32.88	240.8	93.41	351.7	148.8								
1	4	4919	8645	202.7	7746	13979	164.9	6.062	25.16	246.9	74.4	169.9	174.7								
1	5	10.74	31	132.8	26.51	44.25	161.6	2.75	9.813	174.2	1465	5433	147								
1	6	4.223	15.53	217.8	4.74	25.5	154.6	2.128	10.91	222.8	3411	7663	153.6								
1	7	4360	7501	197.2	10278	18284	157.8	35.35	65.78	196.5	121.2	215.9	230.9								
1	8	4185	7740	175.6	10659	20359	134.8	34.35	66.22	211.7	124.9	246	214.8								
1	9	947.2	1829	188.5	326.6	252.1	143.9	3.193	17.72	236.2	4.538	7.219	169.3								
1	10	898	1634	202	7.778	38.97	166.2	3.273	12.09	235.4	3.157	9.625	197.3								
1	11	48.42	87.56	244.7	7.337	28.78	188.2	5519	9657	164.5	28.03	54.75	249.4								
1	12	258.2	480.7	219.4	9.333	25.47	193.9	8231	13124	176.8	170.3	329.9	230								
1	13	5839	10142	208.5	16.04	49.38	211.9	52.88	128.8	217.8	7174	12634	216.9								
1	14	5873	11083	179.8	47.1	97.81	166.8	28.77	57.75	208	5123	9737	189.9								
1	15	85.5	74.28	111.8	5251	9316	162.6	194.2	320.6	223	89.78	59.56	151.4								
1	16	18.34	30.28	213.6	5943	11316	137.1	297.1	517.4	204.3	6.706	10.66	220.3								
2	1	4.337	13.03	180	192.4	314.6	180	1790	3201	211.9	1565	4426	94.23	10014.3	1	1	0	0	0	0	0
2	2	2.306	10.16	155.7	247.9	477.2	206.4	3059	5137	231.8	41.02	94	91.3								
2	3	2547	4615	112.6	4789	9119	135.3	12	31.44	206.2	108.7	346.5	157.2								
2	4	4800	8616	150	7635	13885	173.5	9.216	22.53	215.1	67.55	167.3	184.7								
2	5	11.35	27.13	81.3	19.25	43.63	161.1	1.52	9.531	250.8	1399	5348	155.5								
2	6	4.346	12.88	147.8	7.594	26.19	187.4	0.622	9.594	295.2	3395	7633	162.5								
2	7	4274	7407	143.8	10408	18268	165.8	29.69	63.16	230.4	120.6	215	239.4								
2	8	4146	7651	122.6	10701	20375	143.1	27.23	63.13	224.7	132.8	250.8	219.4								
2	9	925.9	1769	134.3	318.5	250.1	152.3	5.544	20.34	191.8	5.264	6.969	199.8								
2	10	908.4	1646	148.4	16.05	45.44	221.1	1.225	12.34	66.35	3.757	11.38	173.7								
2	11	46.23	89.38	191.7	2.491	23.72	150.4	5618	9808	172.1	32.86	58.47	264.7								
2	12	252	474.7	164.3	1.286	25.5	165.4	8303	13169	184.1	172.8	331.8	240.2								
2	13	5786	10087	155.9	28.18	56.63	198.6	59.94	133	226.4	7139	12607	224.9								
2	14	5881	10996	126.7	31.84	89.81	185.2	43.54	67.59	210.1	5076	9620	198.2								
2	15	85.42	74.63	59.18	5316	9309	170.5	210	343.8	228.6	95.57	63.66	160.3								
2	16	11.78	30.53	171.7	5945	11312	145.3	326.6	540.5	210	3.121	10.69	231.4								

Hardware Maintenance

- Routine testing of fibers, diodes, and PMTs is recommended.
  - Frequency of checks dependent on use
- Using a phantom to acquire sample data (keeping gain constant):
  - Test each fiber in a standard diode
  - Test each detector bundle in a standard PMT
  - Using a “good” fiber (one reserved only for testing), test each diode
  - Using a “good” detector bundle, test each PMT