



Challenges and Lessons Learned
from One Year of X-ALD Newborn Screening
in a Two-Screen State

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Public Health Laboratories
Washington State Department of Health

New Conditions Grant (UC9MC30369)

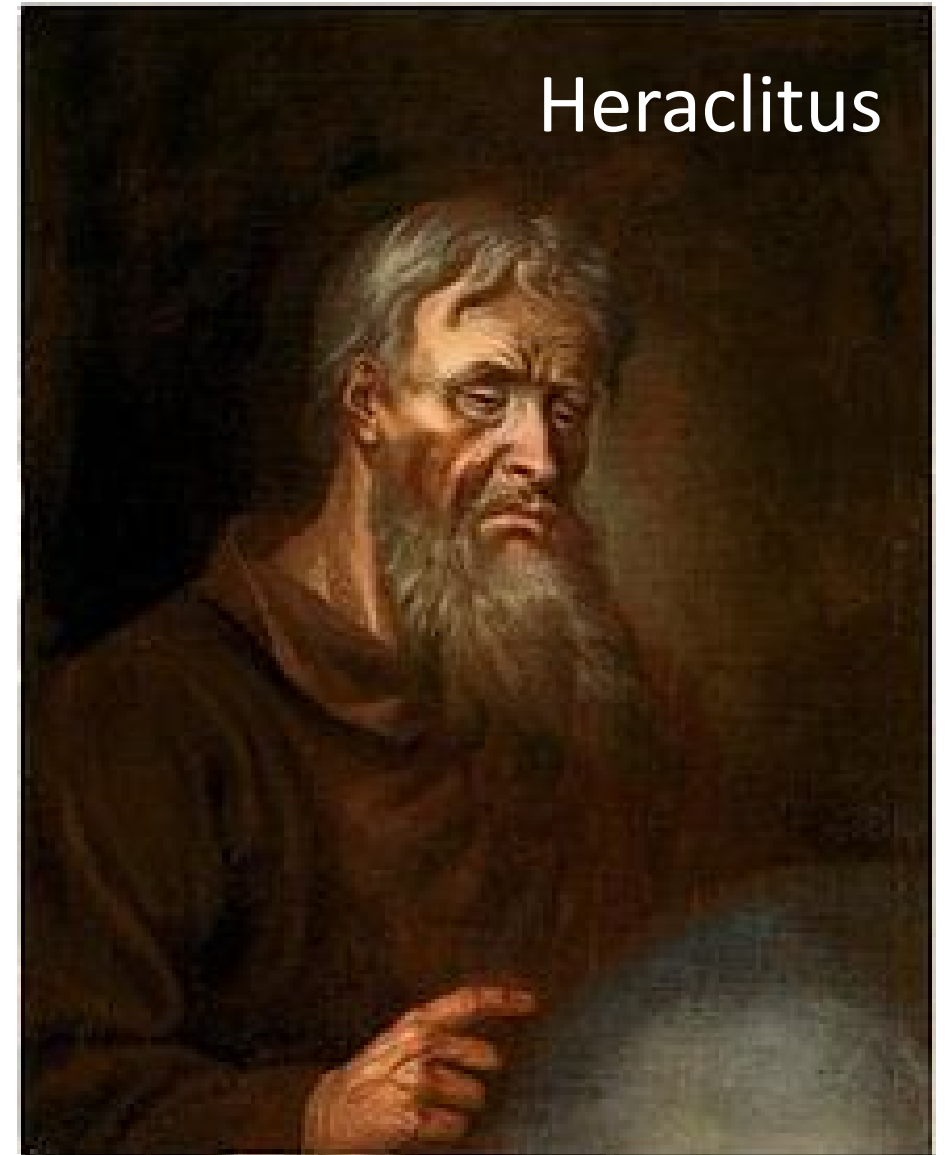
- Method development
- Educational outreach campaign
- Long-term follow-up



Everything changes and
nothing stands still.

...

(The only constant is change.)



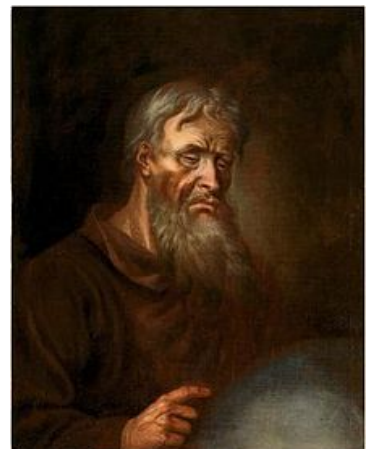
Sequence of Events

- New lab construction
- Purchase instruments
- Validate C26:0 LPC assay
- Hire staff
- Increase fee
- Implement screening



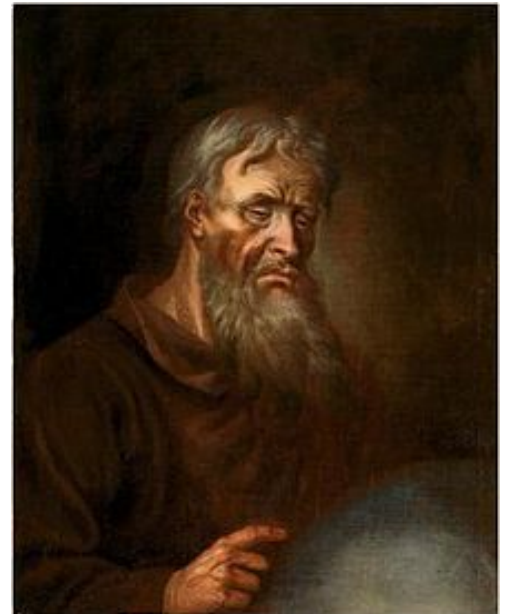
Validate C26:0 LPC assay

- Construction still ongoing
- Started validation in old lab on existing Xevo MS/MS
- Entrance into new MS/MS lab weeks before start date.
- Assay validated (thank you to NY for positive specimens)



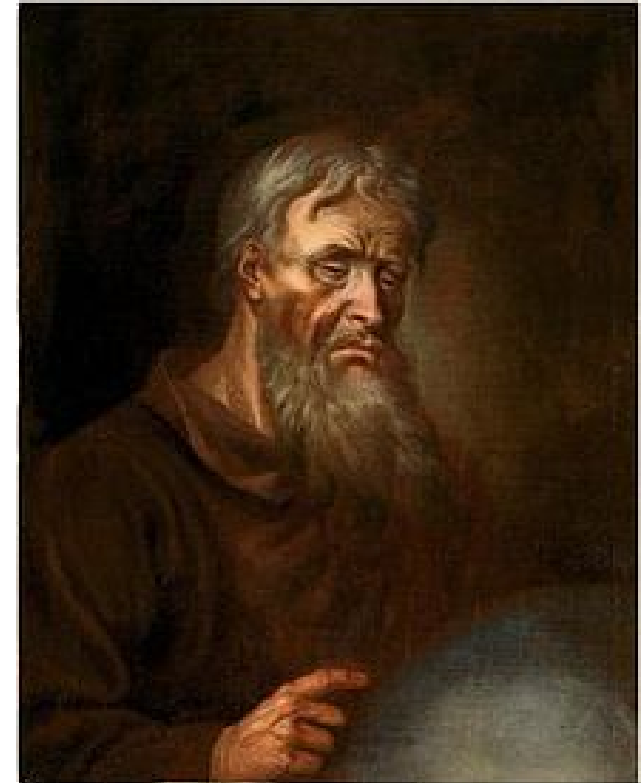
Hires and Fee Increase

- Hiring went smoothly
 - 1 chemist - analyst
 - 1 lead worker
 - 1 follow-up staff
- Fee increase did not go smoothly
 - Requested a \$10/baby increase
 - Legislature gave us \$8.10/baby increase



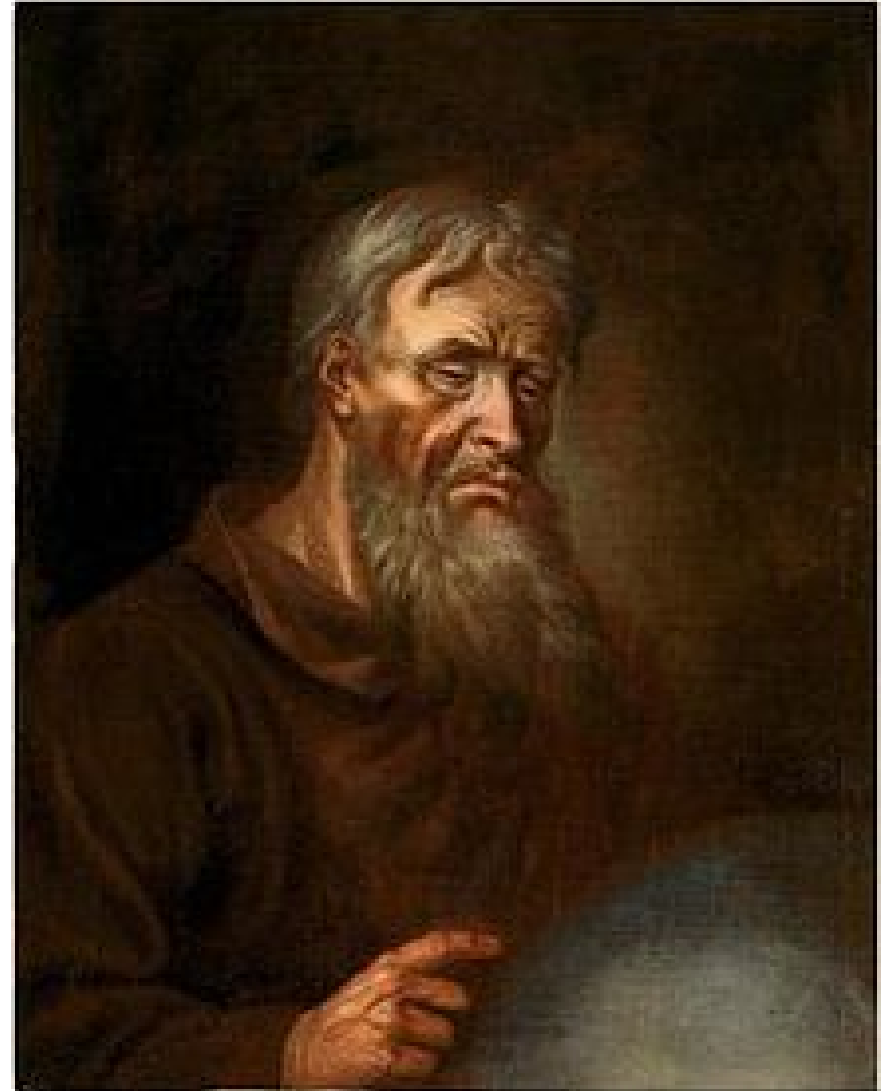
Start of Screening

- Delayed by about three weeks
- Punched specimens into plates and kept them for later analysis
- Catch-up over a period of several weeks
- Decided to print double mailers
 - X-ALD pending message



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C26:0 LPC (lysophosphatidylcholine)

- Initial cutoff: C26:0 LPC < 0.15 umol/L (normal)
 - Phone call to check clinical status
 - If symptomatic of Zelleweger syndrome, refer
 - Otherwise, request repeat screen
- Referrals for persistent elevation of C26:0 LPC
 - Very-long chain fatty acid analysis
 - DNA (if needed)

X-ALD screening results

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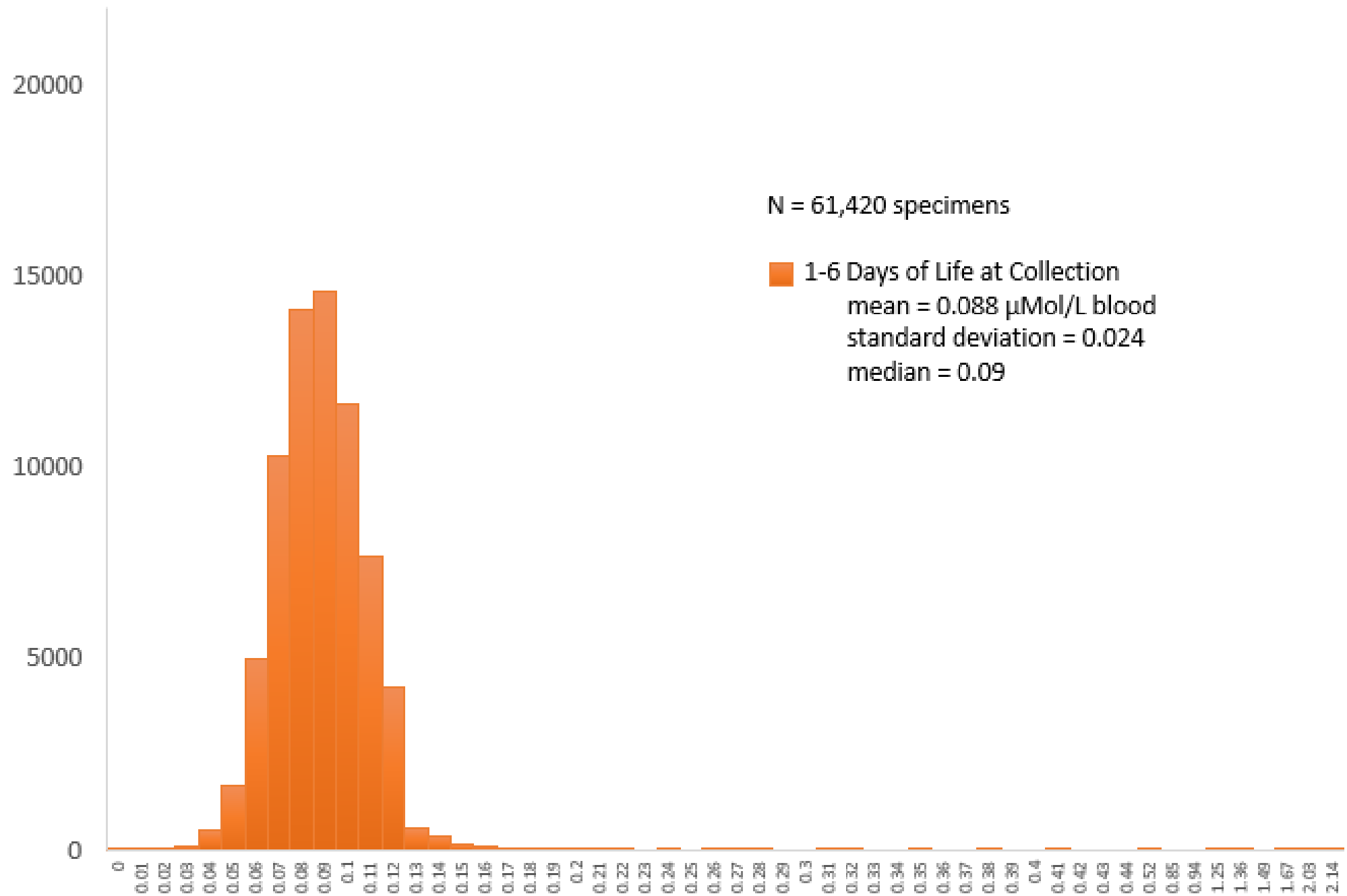
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<ul style="list-style-type: none">VLCFA uncertain (1 ruled out, 1 DNA pending)	2

PPV=33%

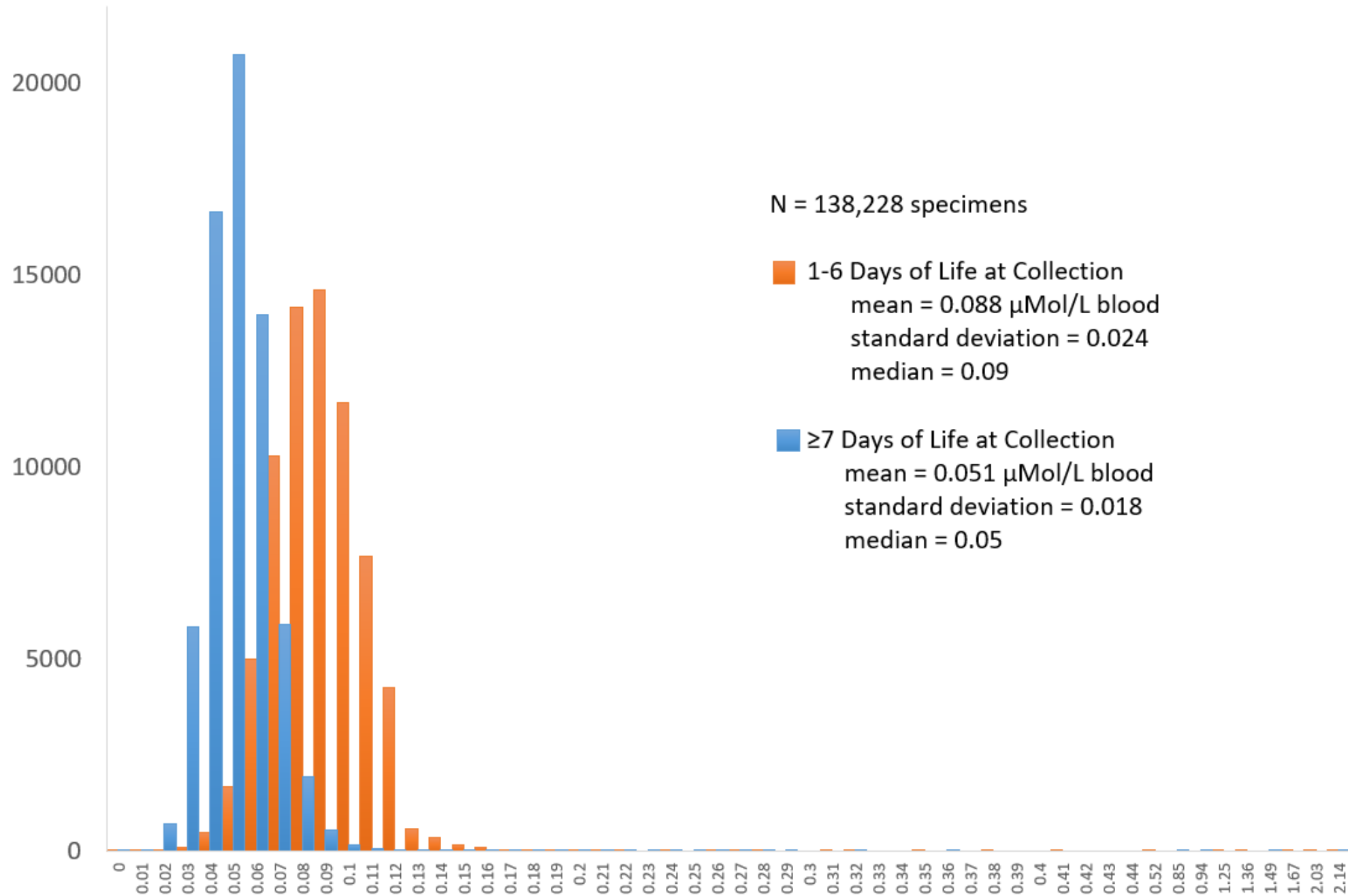
Birth Prevalence

- Expected prevalence
 - 1:14,000 boys
- Observed prevalence
 - 1:11,000 boys
 - 1:7,000 girls
- Zelleweger syndrome
 - 1:21,000 births

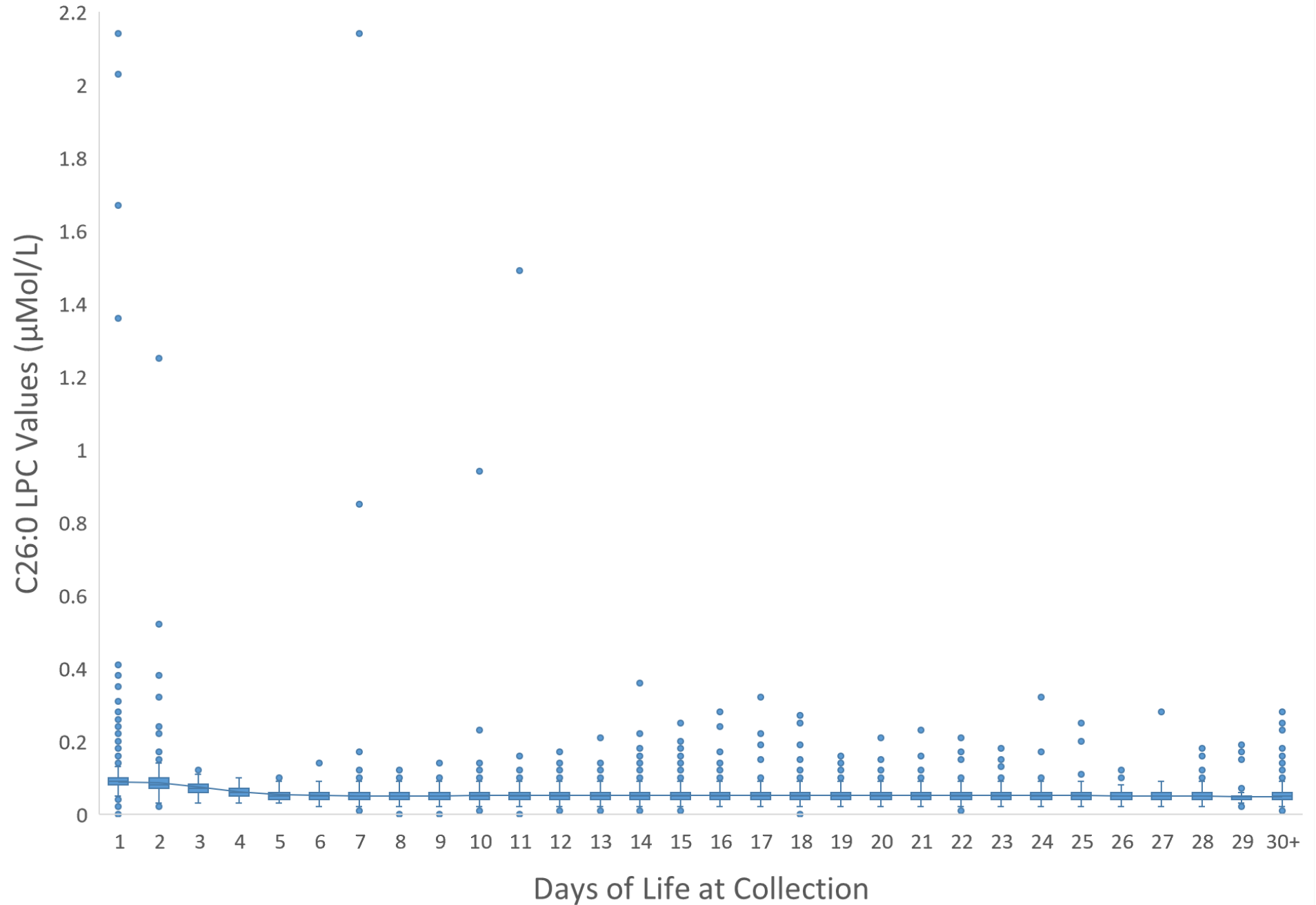
C26:0 LPC Values by Day of Life at Collection



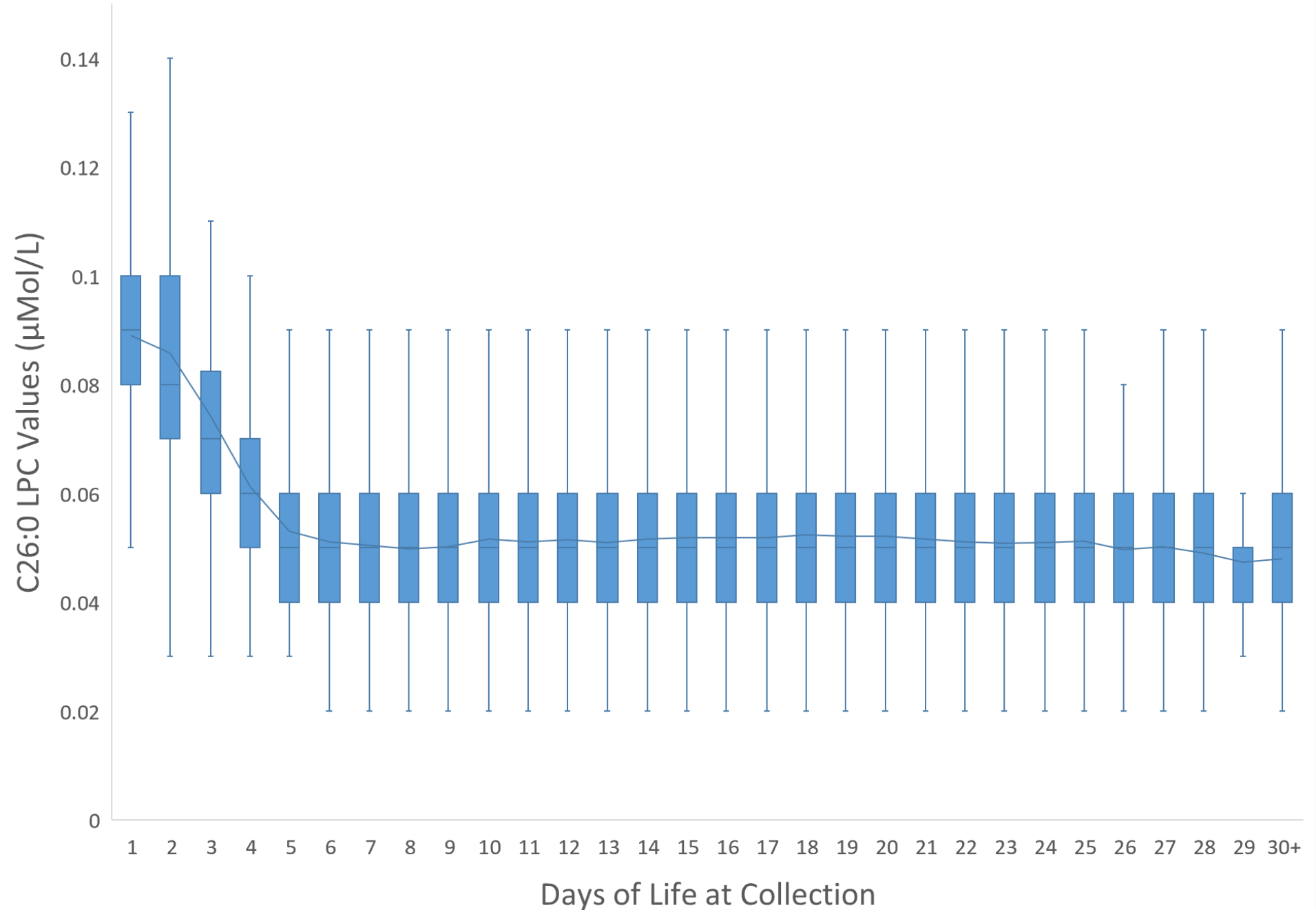
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Clinical partnership/LTFU

- Partnership
 - Biochemical genetics (lead specialists)
 - Endocrinology
 - Neurology
- Increased volume of phone calls to BCG clinic
 - New condition effect?
 - Neurological disorder impact?
- Need resources for LTFU
- IRB approval for LTFU?



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Data Analysis

- Cutoff originally set at 99.98th percentile (0.15)
- Pulled data for period 3/1/2018 through 12/31/2018
- Cleaned data
- Preliminary exploration (relationships between sex, BW, NICU status, AAC with C26:0 LPC values)
 - Histogram of full data set vs. 1st and 2nd DBS
 - Box plots

Data Analysis

- Descriptive statistics (entire data set & <7 DOL vs ≥ 7 DOL)
- Plotted VLCFA interpretations with C26:0 LPC values for specimens screened during that period
- Determined MoM for each abnormal result

Table 1. Referrals under current and proposed C26.0 LPC cutoff

	MoM* 1st	MoM* 2nd	MoM* 3rd	MoM* 4th	VLCHA Interpretation [#]	Sex	Variant	Referral?	
								Original Determination	Proposed Cutoff
Z4	23.72				Zellweger	F		Y	Y
Z3	22.56	42.80			Zellweger	F		Y	Y
Z2	18.50	29.70			Zellweger	F		Y	Y
Z1	13.89	18.80			Zellweger	F		Y	Y
X5	4.29	4.75			X-ALD	M		Y	Y
X4	15.06	17.00			X-ALD	M		Y	Y
X2	4.56	4.60			X-ALD	M		Y	Y
X1	3.89	7.20			X-ALD	M	VOUS	Y	Y
X3	5.72	6.30			X-ALD	F		Y	Y
C4	4.83	5.25			Heterozygote	F		Y	Y
C3	3.44	4.25			Heterozygote	F		Y	Y
C5	6.07	6.50	3.88		Carrier	F		Y	Y
C2	4.22	4.20			Carrier	F		Y	Y
C1	4.22	6.40			Carrier	F		Y	Y
U2	1.79	3.38			Uncertain clinical significance	F		Y	N
U1/N19	1.22	4.00	3.30		Unable to R/O	F	None detected	Y	N
N09	3.44	3.80			Normal	M		Y	Y
N08	1.11	5.10	3.50		Normal	M		Y	Y
N10	0.89	5.50	3.70		Normal	F		Y	Y
N03	1.67	1.20	3.80		Normal	M		Y	uncertain [@]
N16	1.39	3.00	4.25		Normal	M		Y	uncertain [@]
N13	1.00	4.90			Normal	F		Y	uncertain [@]
N11	1.00	5.00			Normal	F		Y	uncertain [@]
N01			3.10	5.60	Normal	F		Y	uncertain [@]
N12	3.56	3.30			Normal	M		Y	N
N05	3.06	3.00			Normal	M		Y	N
N04	3.00	3.30			Normal	M		Y	N
N17	2.00	2.25	2.75		Normal	M		Y	N
N18	1.22	0.40	4.40	2.40	Normal	M		Y	N
N15	1.22	3.88	2.88		Normal	M		Y	N
N07	1.11	4.60	3.30		Normal	M		Y	N
N14	1.22	4.90	3.00		Normal	F		Y	N
N06	1.11	3.00	3.10		Normal	F		Y	N
N02	0.89	4.50	3.00		Normal	F		Y	N

* MoM: multiple of the median for each specimen (values highlighted in blue are at least 3.4 times the median).

VLCHA Interpretation was extracted from the diagnostic lab report.

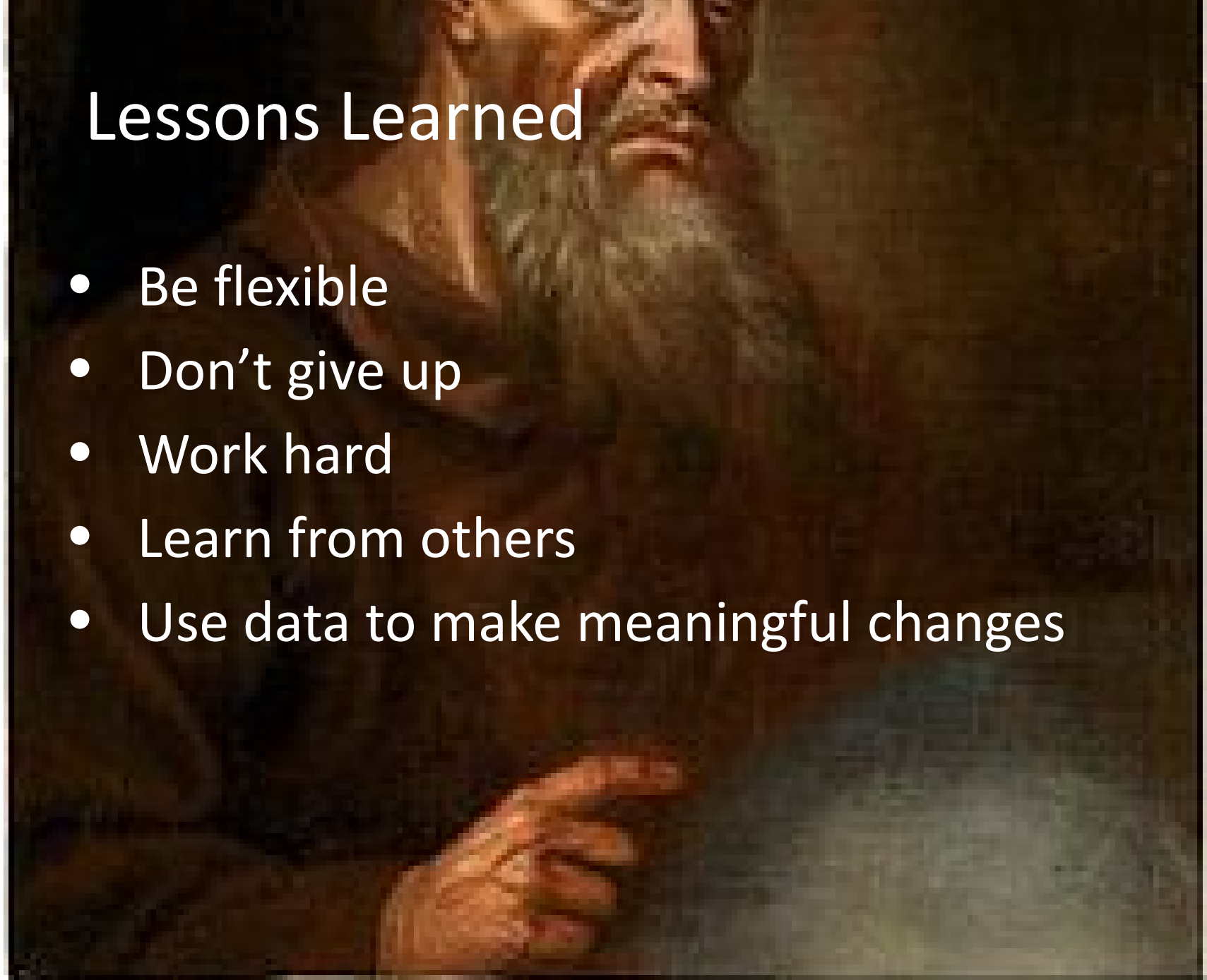
@ It is uncertain if a referral would be recommended in these cases under the proposed cutoff unless a subsequent specimen was also elevated. In some cases, the elevated specimen would not have been collected with the proposed cutoff.

- Proposed new cutoffs at 3.2 x median
 - 0.22 (<7 DOL)
 - 0.12 (≥7 DOL)
- Anticipate
 - reduction in false(+) by close to 80%
 - reduction in number of referrals by about 30%



Lessons Learned

- Be flexible
- Don't give up
- Work hard
- Learn from others
- Use data to make meaningful changes



Acknowledgements

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- * Neurology

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