Evidence-Based Harmonization of Adult Reference Intervals Across Canada using Big Data Analytics: A Report of the CSCC Working Group on Reference Interval Harmonization (hRI)

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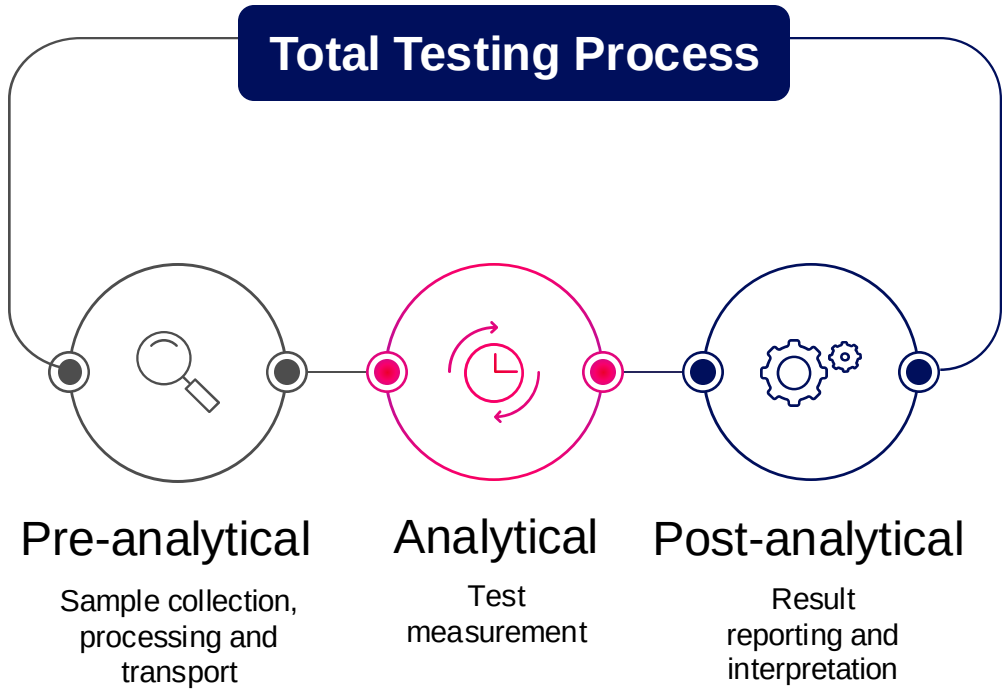
On Behalf of the CSCC Reference Interval Harmonization Working Group

September 27th 2021
AACC Student Abstract Oral Presentation

Harmonization in Laboratory Medicine

- Harmonization is a fundamental aspect of ensuring the analytical and clinical quality of the ***total testing process***
- Growing expectation for standardized patient care across healthcare centers
- Harmonization efforts have largely focused on the pre-analytical and analytical phase of testing, including:
    - *Standardized quality indicator goals*
    - *Increased automation*
    - *Development of commutable reference standards and improved metrological traceability*

Have similar gains been made in reference interval reporting?



Clinical chemistry and laboratory medicine. 2013 Apr 1;51(4):741-51.

CSCC Working Group on Reference Interval Harmonization

Main Objective: Establish evidence-based harmonized/common reference intervals (hRIs) and support their implementation in laboratories across Canada.

Co-Chairs
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Christine Collier

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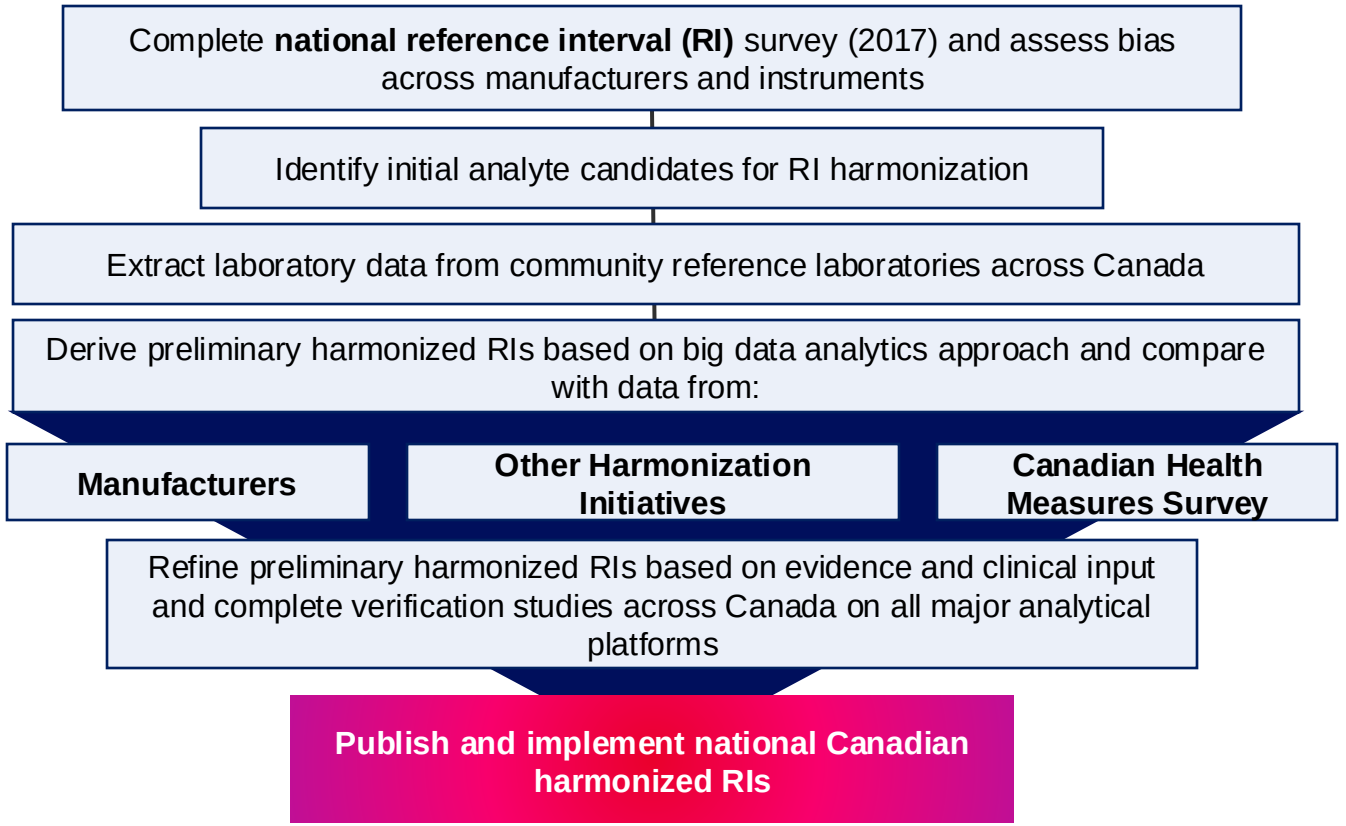


Reference Interval Harmonization in Canada: *Current Gaps*

Reference interval harmonization supports consistent and standardized test result interpretation***, when appropriate***

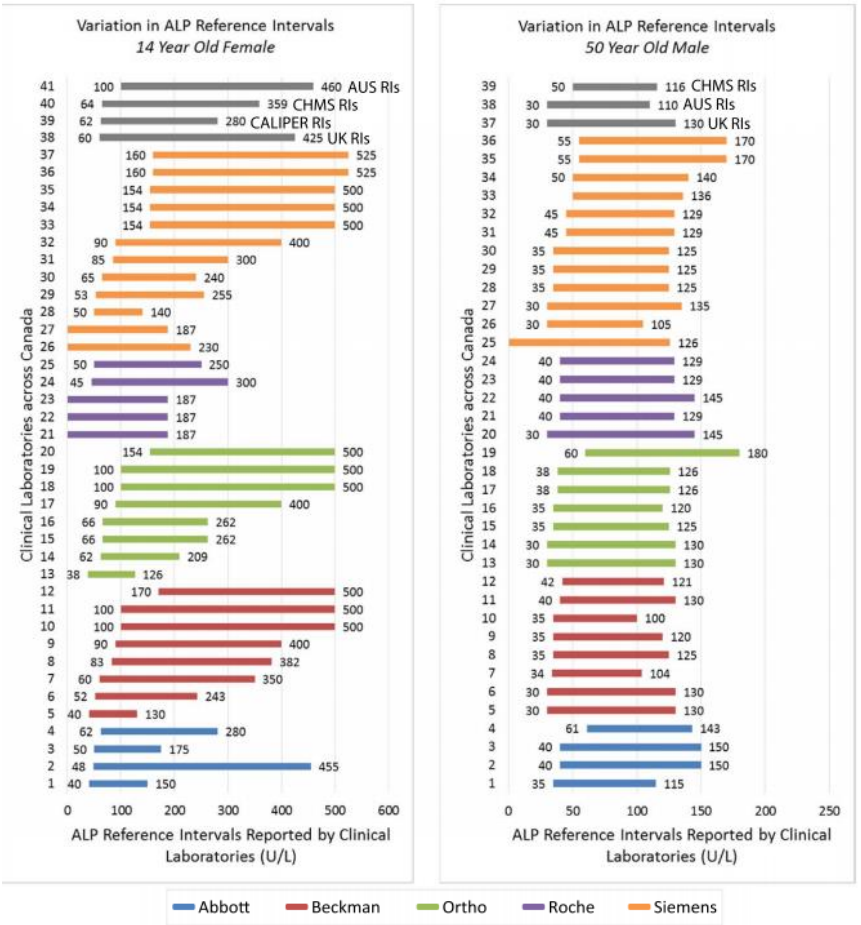
CSCC 2017 National Survey on Reference Interval Variation:

Design:

- 37 laboratories, 7 analytes: RIs for ALT, ALP, calcium, creatinine, fT4, hemoglobin, sodium
- 40 laboratories measured 6 analytes in reference samples (hemoglobin excluded)

Key Findings:

- Variability in RIs even between laboratories using the same instrumentation
- RI variability exceeded test result variability



Clinical biochemistry. 2017 Nov 1;50(16-17):925-35.

CSCC hRI WG: Selection of initial analyte panel

- Candidate analytes for harmonization must demonstrate minimal analytical bias across the platforms to be harmonized

- For the analytical platforms used in Canada, we evaluated:
  - ***Method***
  - ***Manufacturer***
  - ***Calibration traceability***
  - ***Reference method***

Electrolytes

- Sodium
- Potassium
- Magnesium
- Chloride
- CO2

Hepatic

- ALT
- ALP
- Total Protein
- Total Bilirubin
- Albumin
- LDH

Renal

- Creatinine
- Calcium
- Phosphate

Endocrine

- Free T3
- Free T4
- TSH

17 initial analytes were selected

CSCC hRI WG: Selection of reference interval approach

Direct Approach

+

- Recommended by CLSI
- Better representation of a healthy population
- Minimal pre-analytical variation

-

- Extensive resource requirements
- Large sample size required
- Updating recommendations as new analytical platforms develop is challenging

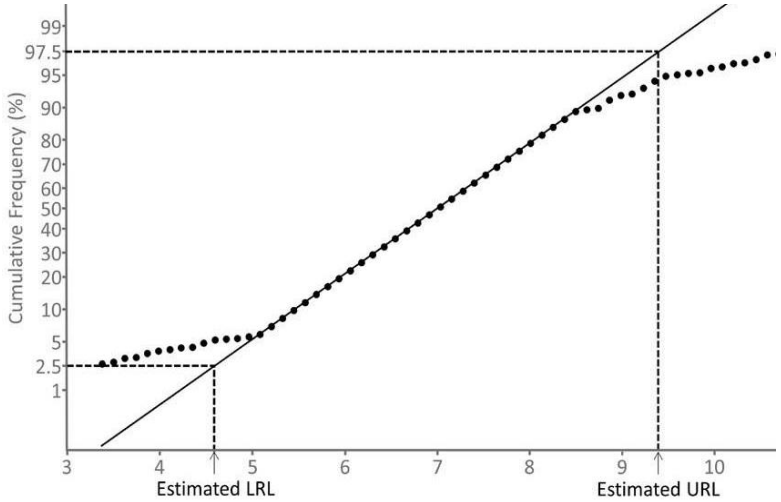
Indirect Approach

- Less resources required
- Data easily representative
- Pre-analytical processes reflect routine laboratory practice

- Requires in-depth statistical analysis and consideration
- Determination of healthy population relies on statistical methods

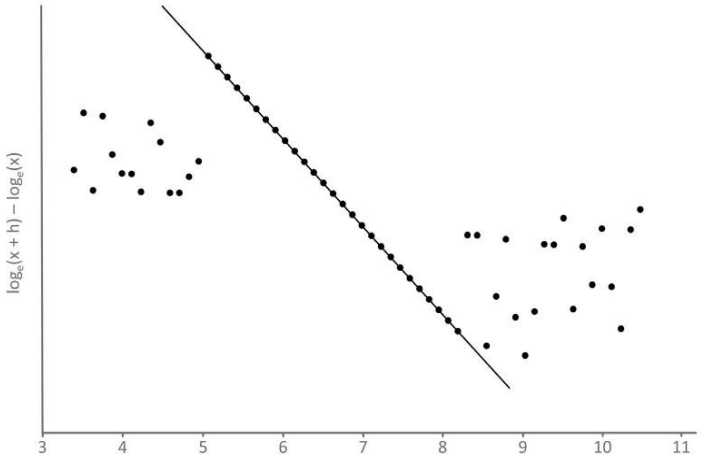
CSCC hRI WG: Selection of reference interval approach

Hoffman Method (1963)



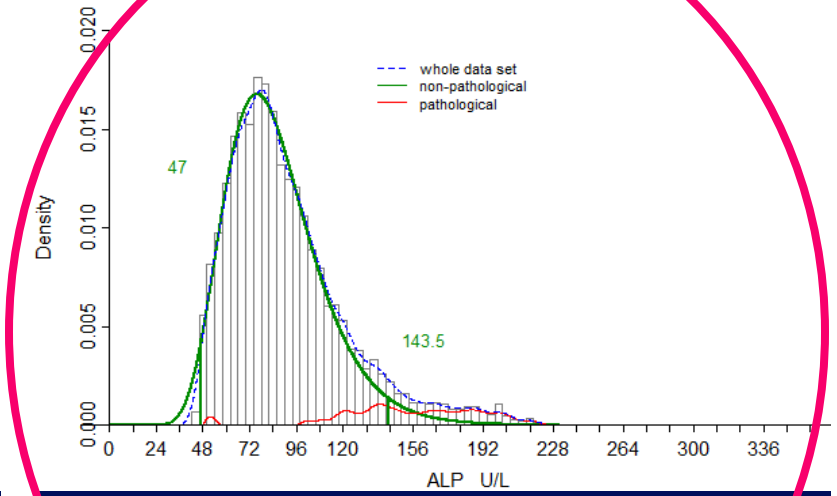
- *Plot the cumulative frequency of the distribution on a normal probability paper*
- *Reference interval extrapolated through linear regression*

Bhattacharya Method (1967)



- *Mathematical straightening of the Gaussian distribution*
- *The slope and intercept are used to determine the mean and SD, and from this, the reference interval*

TML Method (2007)



- *Modern computational power can be leveraged to derive indirect reference intervals using "maximum likelihood estimation"*

The Clinical biochemist Reviews. 2019 May;40(2):99.

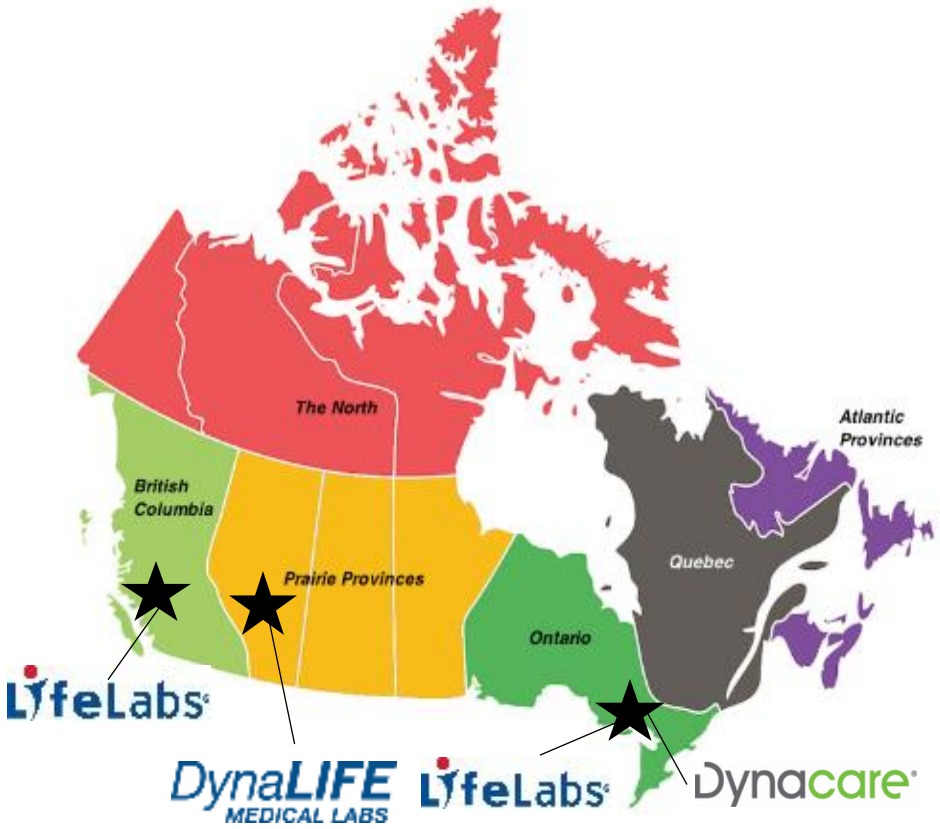
CSCC hRI WG: Selection of data contributing centres

- Appropriate selection of data contributing centres is essential to optimize the performance of indirect methods

Criteria for data centre contribution:

- *Large outpatient population*
- *Representative of Canadian population*
- *Representative of different analytical platforms*
- *Consistent results over time*

Formed collaborations with community laboratories to support this initiative



Analyzing the Data:

Example – Alkaline Phosphatase

1 ***Retrieve population dataset***

- ✓ **Extract data from multiple centres across two year period**
- ✓ **Remove all repeat observations**
- ✓ **Include key covariates:**
  - **Age**
  - **Sex**
  - **Date of Collection**
  - **Result**

Dynacare®

Province: Ontario
Analytical Platform: Roche
Sample Size: 1062848

DynaLIFE MEDICAL LABS

Province: Alberta
Analytical Platform: Siemens
Sample Size: 503169

LifeLabs®

Province: Ontario
Analytical Platform: Roche
Sample Size: 2655240

LifeLabs®

Province: BC
Analytical Platform: Roche
Sample Size: 781171

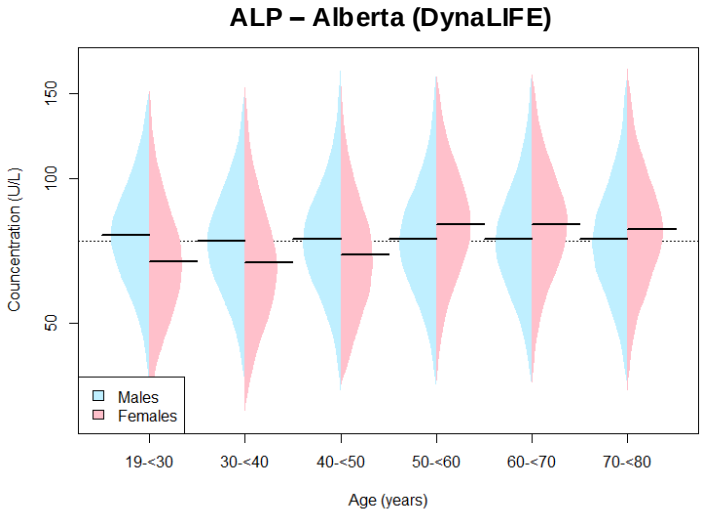
AAC CSCC hRI WG

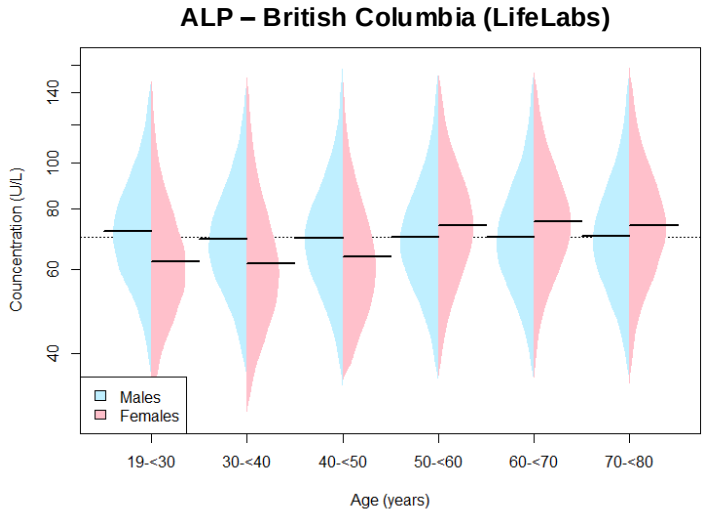
Analyzing the Data:

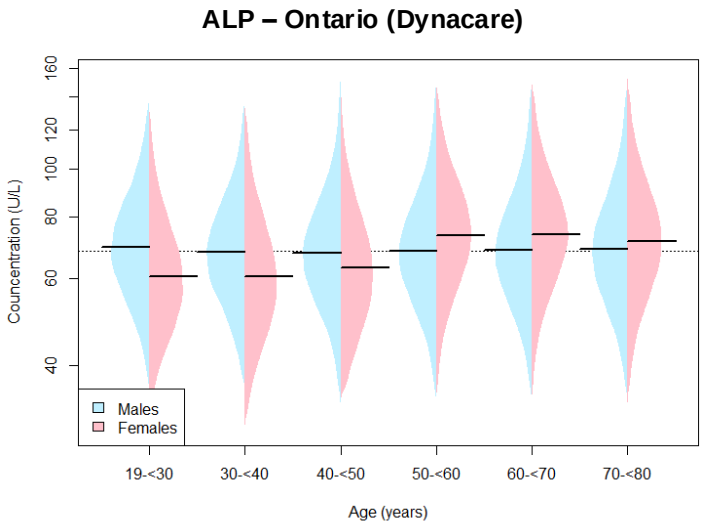
Example – Alkaline Phosphatase

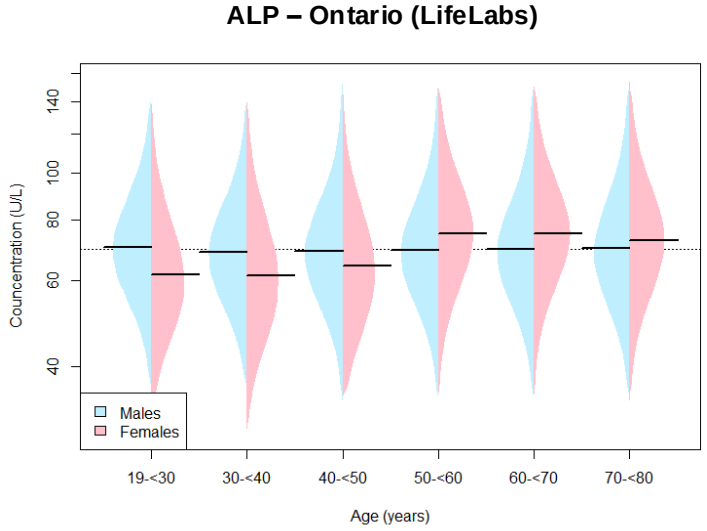
2 ***Assess age/sex differences***

- ✓ **Visually assess raw data across each centre**
- ✓ **Assess data density to evaluate age-specific trends**
- ✓ **Use specialized plots to view age- and sex-specific differences**
- ✓ **Confirm visual assessment statistically using Harris & Boyd Method**









Significant age- and sex-specific differences (20-40y M/F, 40-80y)

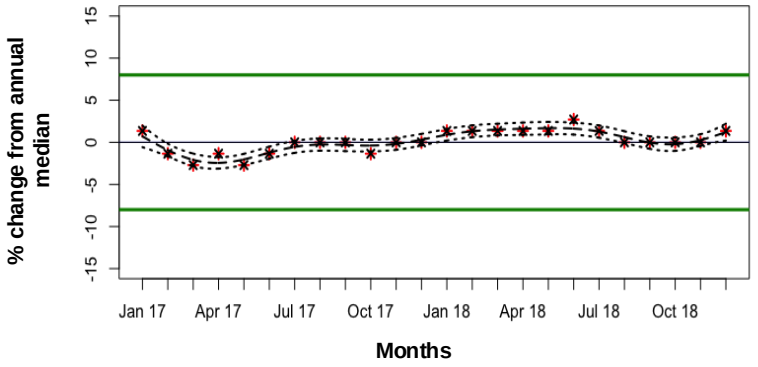
Analyzing the Data:

Example – Alkaline Phosphatase

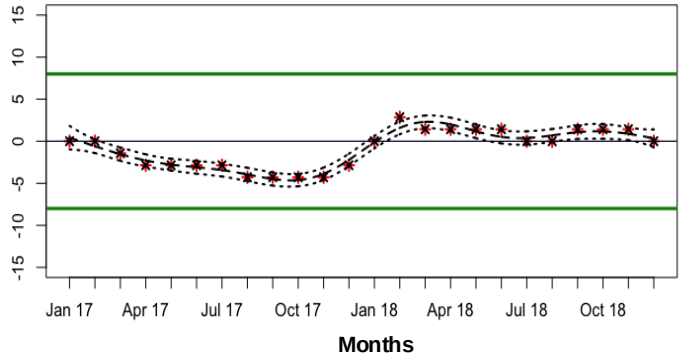
3 ***Data clean up***

- ✓ *Monthly stability assessed visually*
- ✓ *Percent deviation from median compared to reference change value (RCV) reported by EFLM*
- ✓ *Remove outliers for each centre based on Tukey or Hubert method*

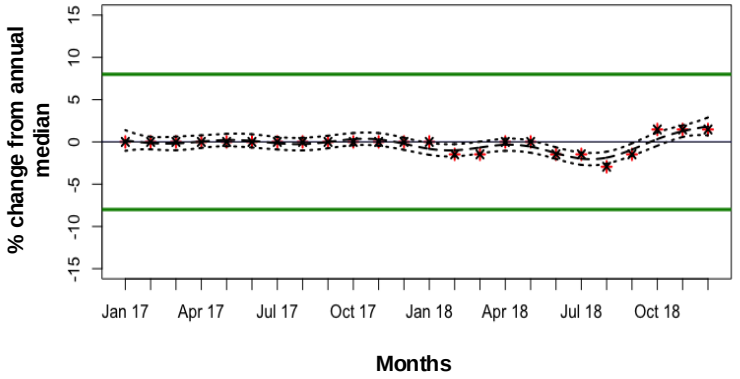
ALP – Alberta (DynaLIFE)



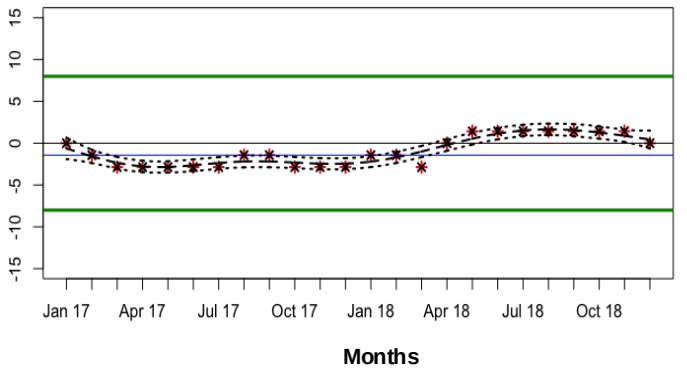
ALP – British Columbia (LifeLabs)



ALP – Ontario (Dynacare)



ALP – Ontario (LifeLabs)



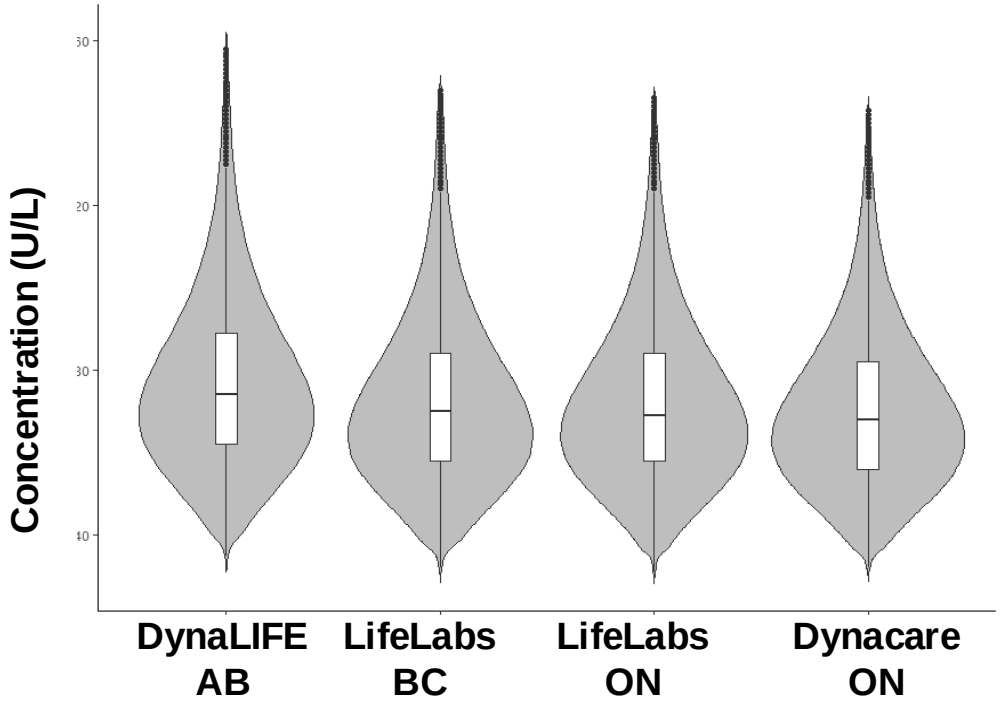
No instability observed

Analyzing the Data:

Example – Alkaline Phosphatase

4 ***Centre-specific differences***

- ✓ *Assess centre-specific differences using Harris & Boyd method*
- ✓ *Combine all centres if no significant differences are observed into Canada-Wide file*



Dynacare®
Province: Ontario (Roche)
Sample Size: 1062848

DynaLIFE MEDICAL LABS
Province: Alberta (Siemens)
Sample Size: 503169

LifeLabs®
Province: Ontario (Roche)
Sample Size: 2655240

LifeLabs®
Province: British Columbia (Roche)
Sample Size: 781171

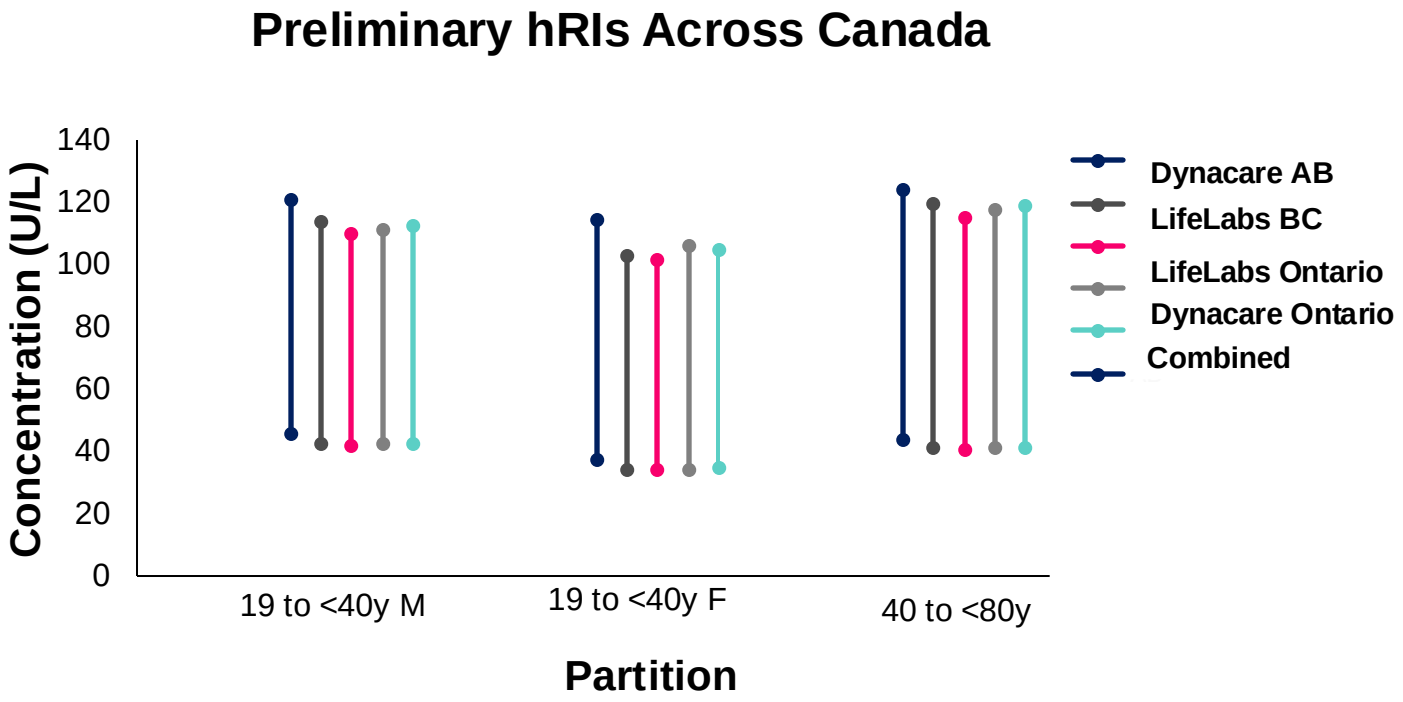
No centre-specific differences

Analyzing the Data:

Example – Alkaline Phosphatase

5 ***Establish RI for each partition***

- ✓ ***Use TML method to establish reference intervals for each partition***
- ✓ ***Compare established reference intervals across provinces and reference intervals***



Partition	Dynacare AB Siemens	LifeLabs BC Roche	LifeLabs ON Roche	Dynacare ON Roche	Canada-Wide
19 to <40y M	46-121	42-114	42-110	42-111	42-113
19 to <40y F	37-115	34-103	34-101	34-106	35-105
40 to <80y	44-124	41-119	41-115	41-118	41-119

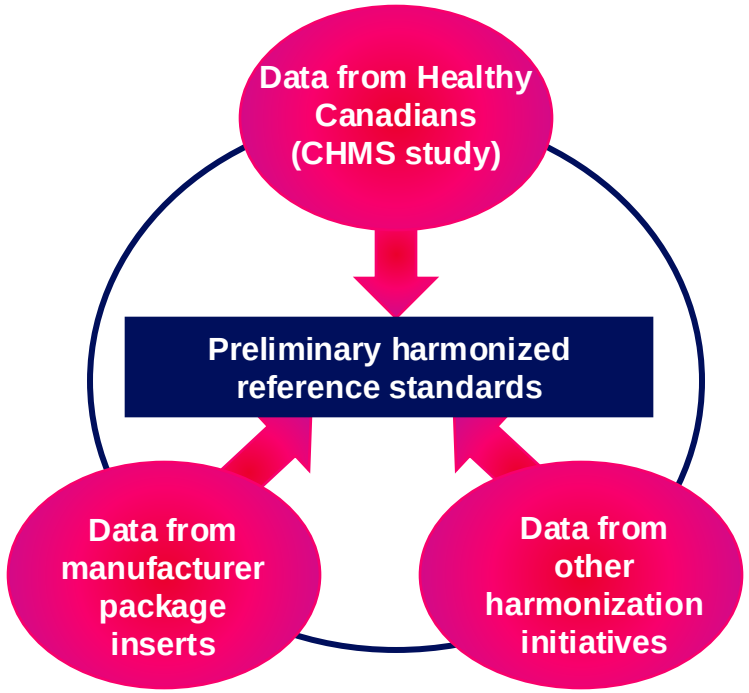
*displayed in U/L

Analyzing the Data:
Example – Alkaline Phosphatase

5 ***Compare and assess***

- ✓ *Compare to indirect and direct data published by international initiatives*
- ✓ *Compare to manufacturer package insert data*
- ✓ *Compare to what is currently used at each centre*
- ✓ *Internal discussion and finalization*

Preliminary Recommendations



Proposed Harmonized RI	
19 to <40y M	40-115 U/L
19 to <40y F	35-105 U/L
40 to <80y	40-120 U/L

CSCC hRI WG: Preliminary Recommendations & Next Steps

Recommended harmonized reference intervals for 17 assays discussed by hRI Working Group

Analyte	Calculated RI		Recommended Harmonized RI	
Alanine aminotransferase (U/L)	19 to <80y M	11-53	19 to <80y M	**RI: 11-53, CDL: <33[a]**
	19 to <80y F	8-35	19 to <80y F	**RI: 8-35, CDL: <25[a]**
Albumin BCG (g/L)	19 to <60y M	40-51	19 to <80 years	**40-50**
	19 to <60y F	39-49		
	60 to <80y	39-49		
Alkaline Phosphatase (U/L)	19 to <40y M	42-114	19 to <40y M	**40-115**
	19 to <40y F	34-103	19 to <40y F	**35-105**
	40 to <80y	41-119	40 to <80y	**40-120**
Lactate Dehydrogenase (U/L)	19 to <80y	122-237	19 to <80 y	**120-240**
Total Bilirubin (umol/L)	19 to <80y M	3.5-20.0	19 to <80 y M	**3-20**
	19 to <80y F	2.8-15.8	19 to <80 y F	**3-16**
Total Protein (g/L)	19 to <80y	61-79	19 to <80y	**60-80**
Phosphate (mmol/L)	19 to <60y	0.79 – 1.45	19 to <80y	**0.80– 1.45**
	60 to <80y M	0.77 – 1.43		
	60 to <8 y F	0.86 – 1.47		
Calcium (mmol/L)	19 to <40y M	2.21 - 2.54	19 to <80y	**2.15 – 2.55**
	19 to <40y F	2.16 – 2.50		
	40 to <80y	2.16 – 2.52		
Creatinine (umol/L)	19 to <80 years M	63-117	*Not finalized*	*Not finalized*
	19 to <80 years F	48-95		
FT3 (pmol/L)	19 to <80y	3.01 – 5.68	19 to <80y	**3.0 to 5.7**
FT4 (pmol/L)	19 to <80y	9.7 - 15.5	19 to <80y	**9.5 to 15.5**
TSH (mIU/L)	19 to <80y	0.60-4.55	19 to <80y	**RI: 0.60-4.55, CDL: 0.1-4.12[b]**
Sodium (mmol/L)	19 to <80y	138-145	19 to <80y	**137-145**
Potassium (mmol/L)	19 to <80y	3.8-5.1	19 to <80y	**3.8-5.1**
Magnesium (mmol/L)	19 to <80y	0.73-1.00	19 to <80y	**0.73-1.00**
Total CO2 (mmol/L)	19 to <80y	22-32	19 to <80y	**22-30**
Chloride (mmol/L)	19 to <80y	97 – 107	*Not finalized*	*Not finalized*

Establishment of preliminary hRIs for 15/17 parameters

Limitations to the current data:

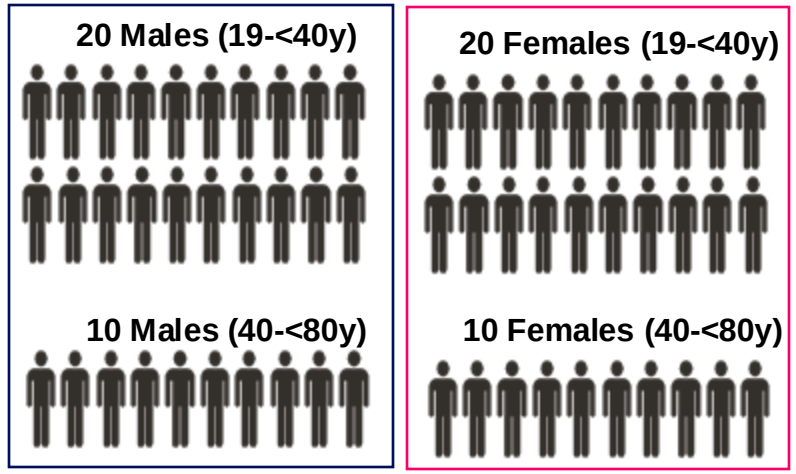
- *Only three manufacturers represented*
- *Only three provinces represented*
- *All data contributing centres use serum as preferred matrices*

How can they be addressed prior to implementation?

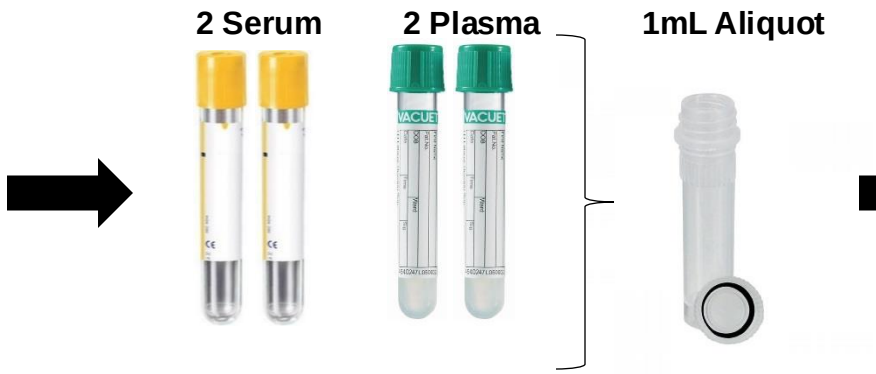
CSCC hRI WG: Cross-Canadian Verification Study

Objective: To verify proposed hRIs on major analytical platforms across Canada using serum and plasma samples prospectively collected from healthy adults.

Study Design:



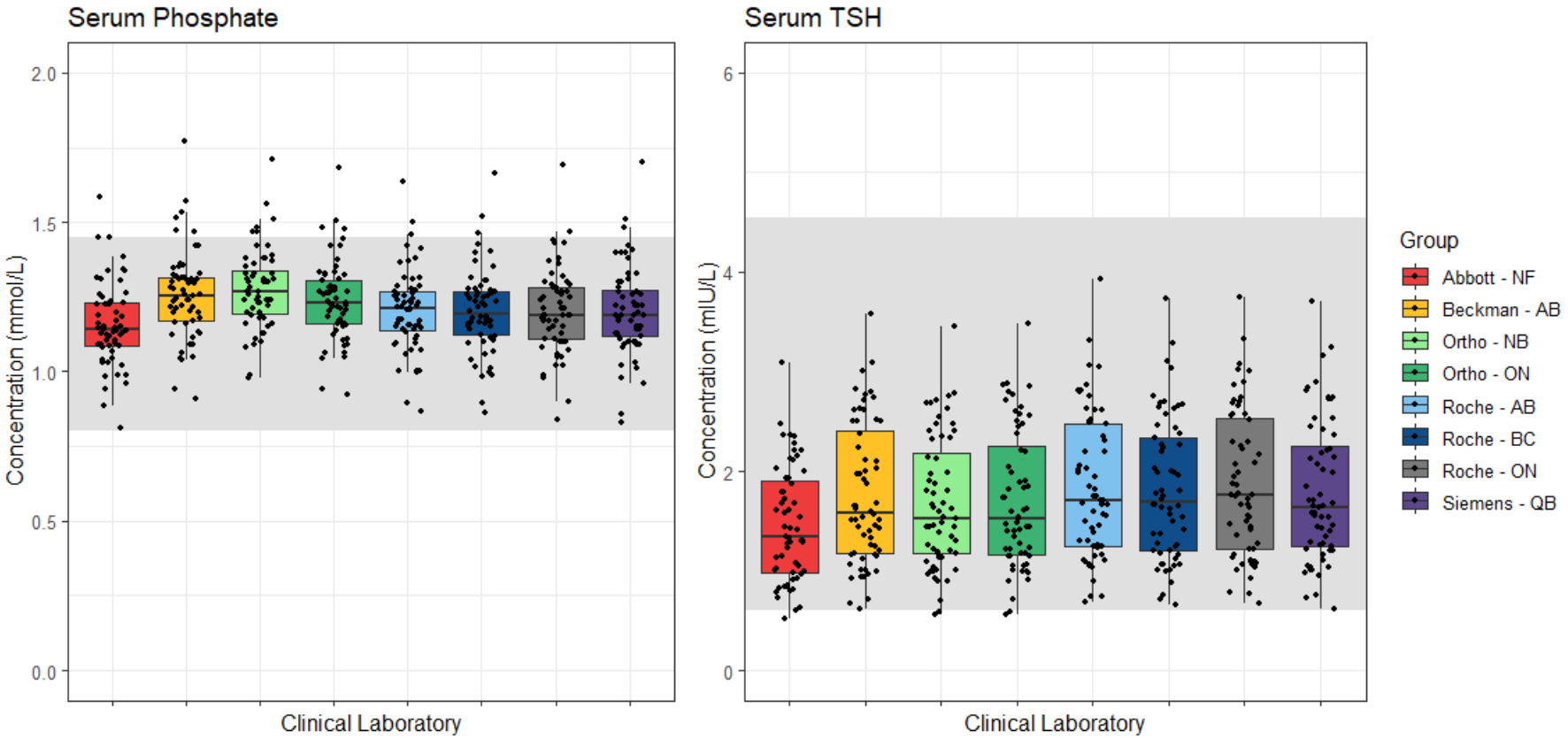
60 adult volunteers *recruited from the community with the following age/sex distribution.*



2 serum and 2 plasma tubes will be collected from each participant

Distribution to 8 laboratories across Canada for analysis of all 17 analytes

CSCC hRI WG: Cross-Canadian Verification Study - Results



- Most analytes met the criteria for verification established by the hRI WG
- Select analytes require further analysis and investigation (e.g. TSH, FT3, FT4, creatinine)
- Minimal differences between matrices were observed

**shaded grey area indicates proposed hRIs*

Final Conclusions and Next Steps

- A **novel big data analytics** approach was undertaken to defined preliminary hRIs for 17 analytes:
  - (1) extraction of data from community reference laboratories across Canada
  - (2) assessment of outliers
  - (3) statistical evaluation of age, sex, and center-specific differences
  - (4) derivation of preliminary hRIs using the TML method
  - (5) comparison of established hRIs to direct data in the healthy Canadian population.

- Robustness of these data was assessed through a **Cross-Canada Verification Study where results supported implementation of these recommendations** (exceptions include: FT4, TSH, FT3, and creatinine)

- Future work will focus on finalizing recommendations, supporting their implementation, and expanding this approach to other analytes

Acknowledgments

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