COMPARABILITY OF METHODS AND ANALYSERS

Nora Nikolac

Zagreb, October 24-25, 2015

15th EFLM Continuing Postgraduate Course in Clinical Chemistry and Laboratory

When?

Introducing new method or analyzer

Multiple analytical systems in laboratory

Using services of another laboratory

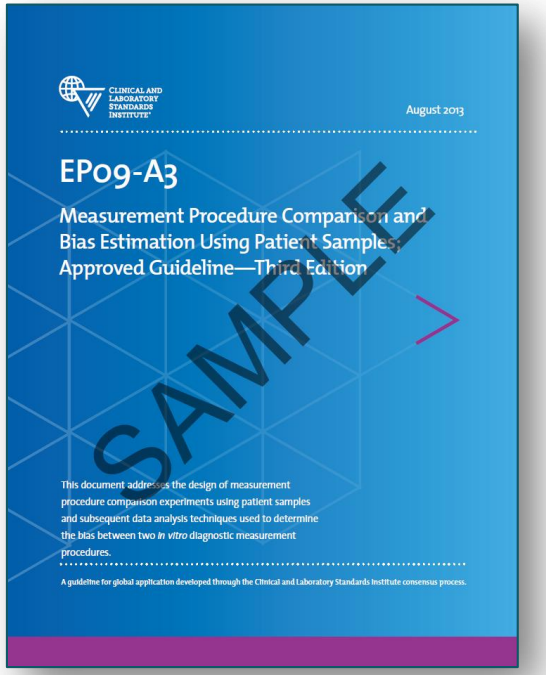
Why?

- To increase patient safety
  - To assure that method change is not going to influence laboratory result for the patient.

How?

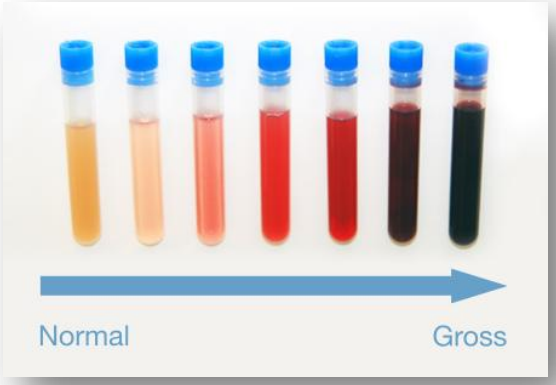
- Experimental procedures following protocols
- CLSI EP09-A3: Measurement procedure comparison and bias estimation using patient samples

1. Number of samples
2. Measurement range
3. Time of analysis
4. Data analysis
5. Data interpretation



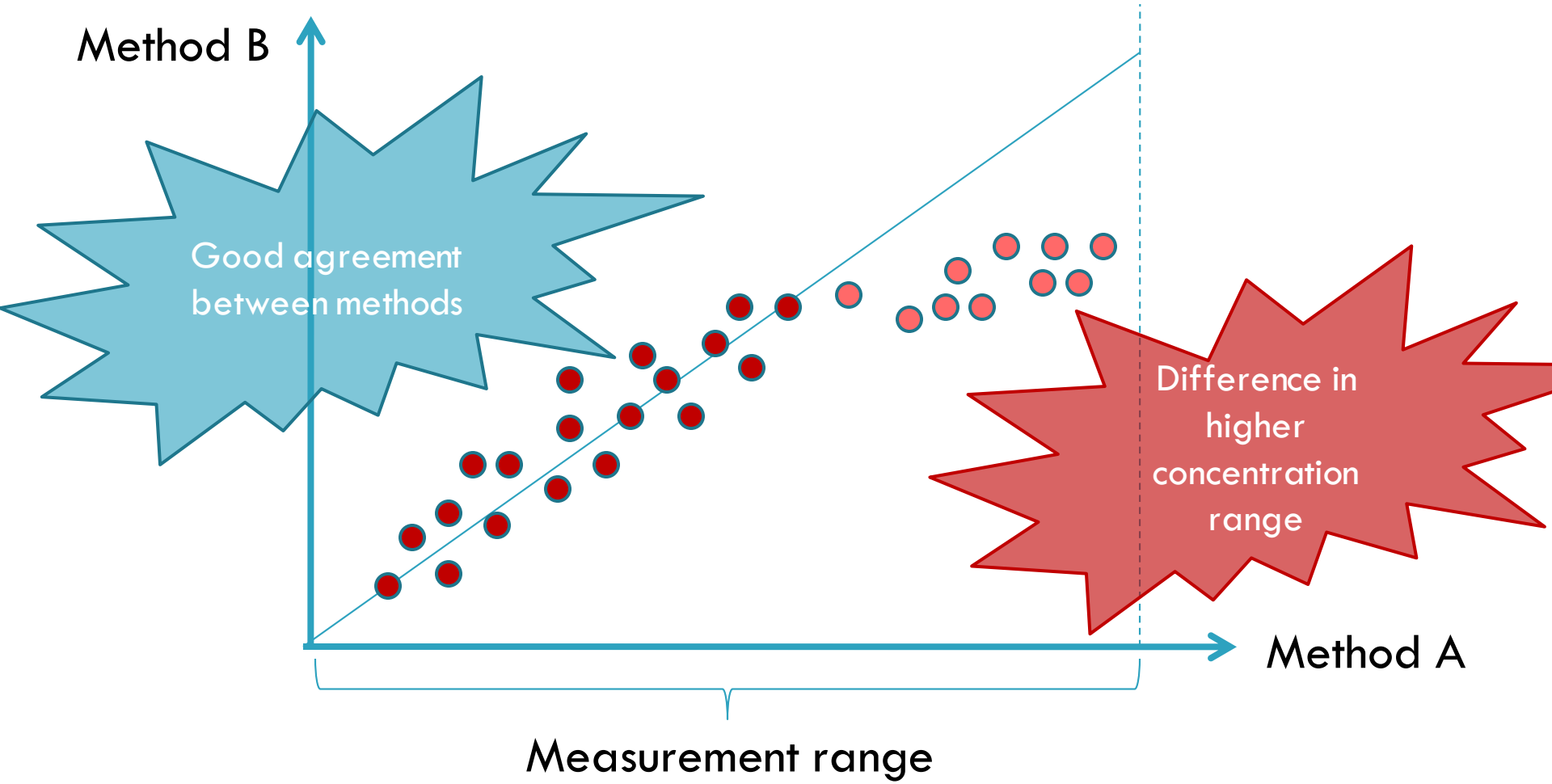
1. Number of samples

- Min: 40 samples
- Optimal: 100 samples
  - To identify unexpected errors from sample matrix or interferences
- Measurements in duplicate



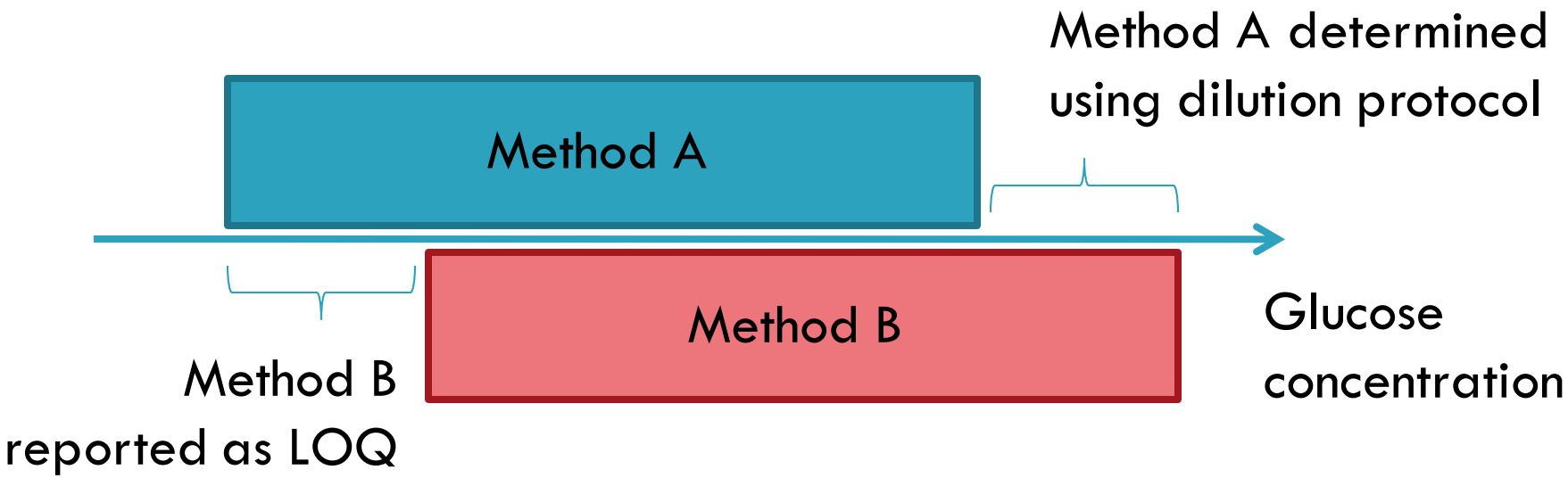
2. Measurement range

- Cover 90% of the method measurement range



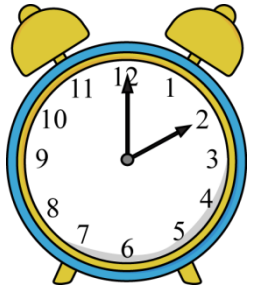
2. Measurement range

- Overlaping measurement range for both methods

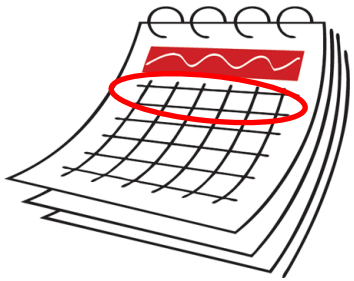


3. Time of analysis

- Measurements done within **2** hours
  - Not for: glucose, lactate, ammonia, blood gass testing...



- Measurements done over **5** days
  - Better over longer period of time

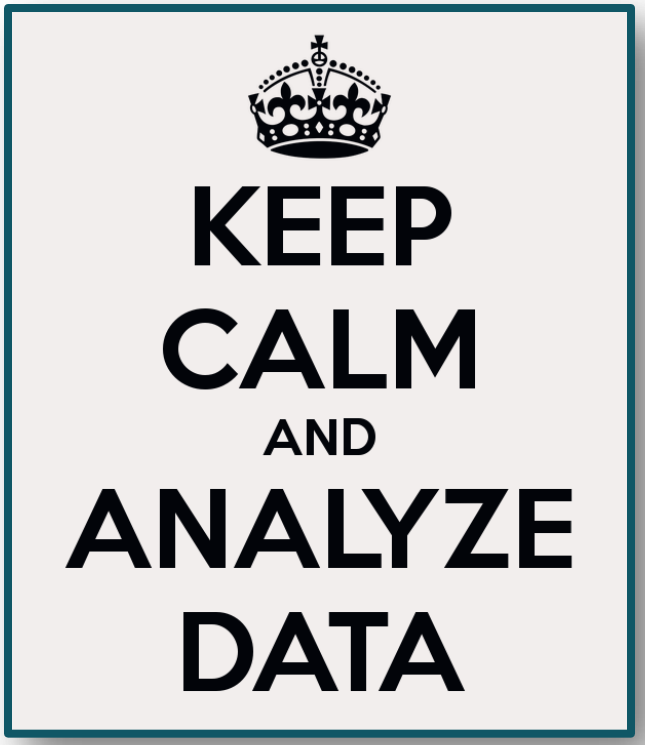


- Collecting samples over period of time (first method) and analyzing in batch using second method

4. Analyzing results

Several statistical aproaches:

- Correlation
- Paired test for difference
- Linear regression
  - Deming regression
  - Passing-Bablok regresion
- Bland-Altman analysis



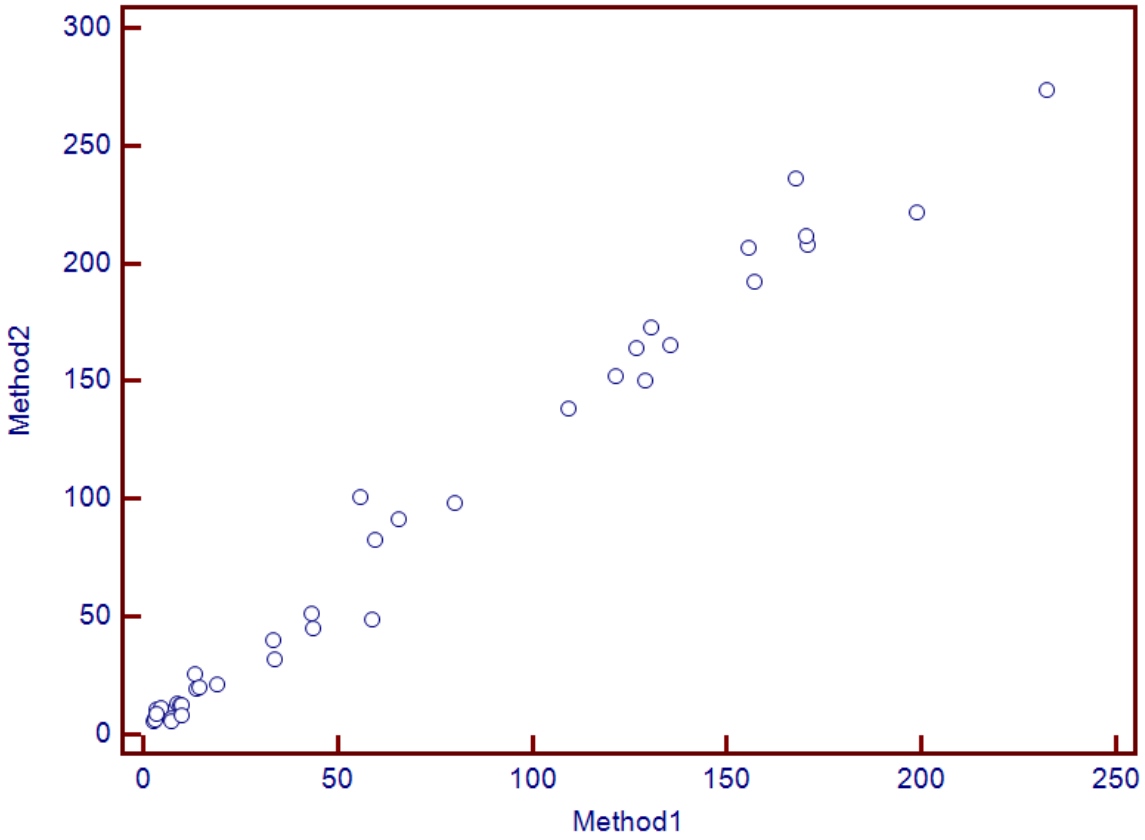
4. Analyzing results

- Comparison of two methods for direct bilirubin concentration measurement

Summary data		
	Method 1 N=40	Method 2 N=40
Analyzer, method	Architect (Abbott) Diazo method	AU 680 (Beckman Coulter) DPD method
Min-Max	2.7-232.3	5.5-273.4
Mean ± SD	65.5 ± 67.9	82.4 ± 83.6
Median (IQR)	38.5 (7.9-127.8)	42.4 (11.1-158.2)
P (normality)	0.059	0.036

4.1 Correlation

- Spearman coefficient of correlation

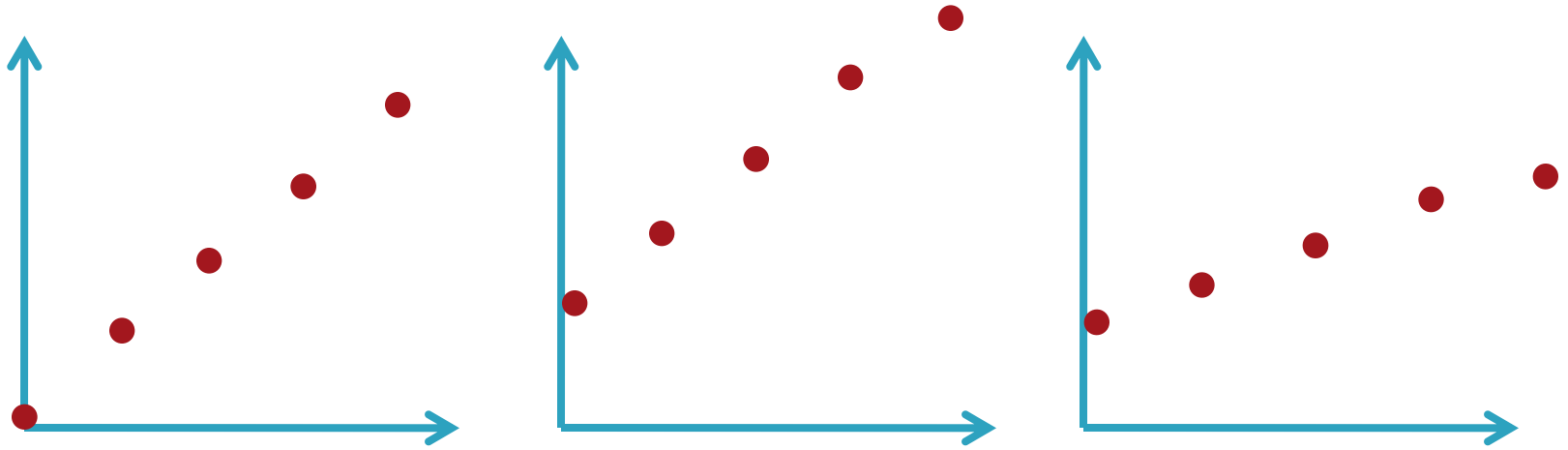


r (95% CI) =
0.97 (0.95-0.98)

Excellant correlation

What is the meaning of this result?

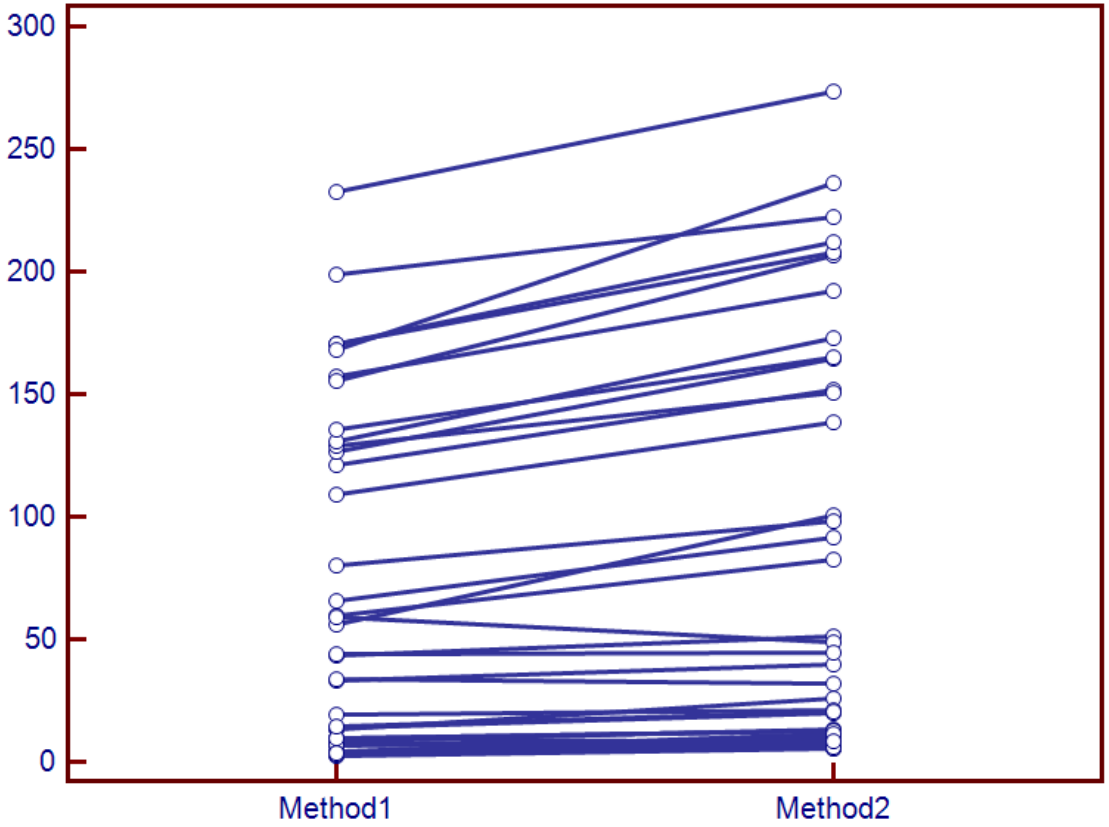
- Methods are significantly associated
- Linear relation between methods
- ↑ of Method A associated with ↑ of Method B
- Nothing about amount of increase!



Same correlation coefficient!

4.2 Significance of difference

- Wilcoxon test (normality failed)



P < 0.001

Significant difference between methods

What is the meaning of this result?

- Calculating differences for each pair of measurement
- Comparing number of negative and positive differences
- If there is no difference between methods, number of differences is equal

Wilcoxon test (paired samples)

Number of positive differences	35
Number of negative differences	4
Large sample test statistic Z	-5,023799
Two-tailed probability	P < 0,0001

Dot-and-Line diagram

More measurements were higher using Method 2

4.3 Linear regression

High correlation
Linear relationship

Equation to describe relationship between methods
Determine proportional and constant error

Deming regression
Passing and Bablok regression

Lessons in biostatistics

Comparison of methods: Passing and Bablok regression

Lidija Bilić-Zulle

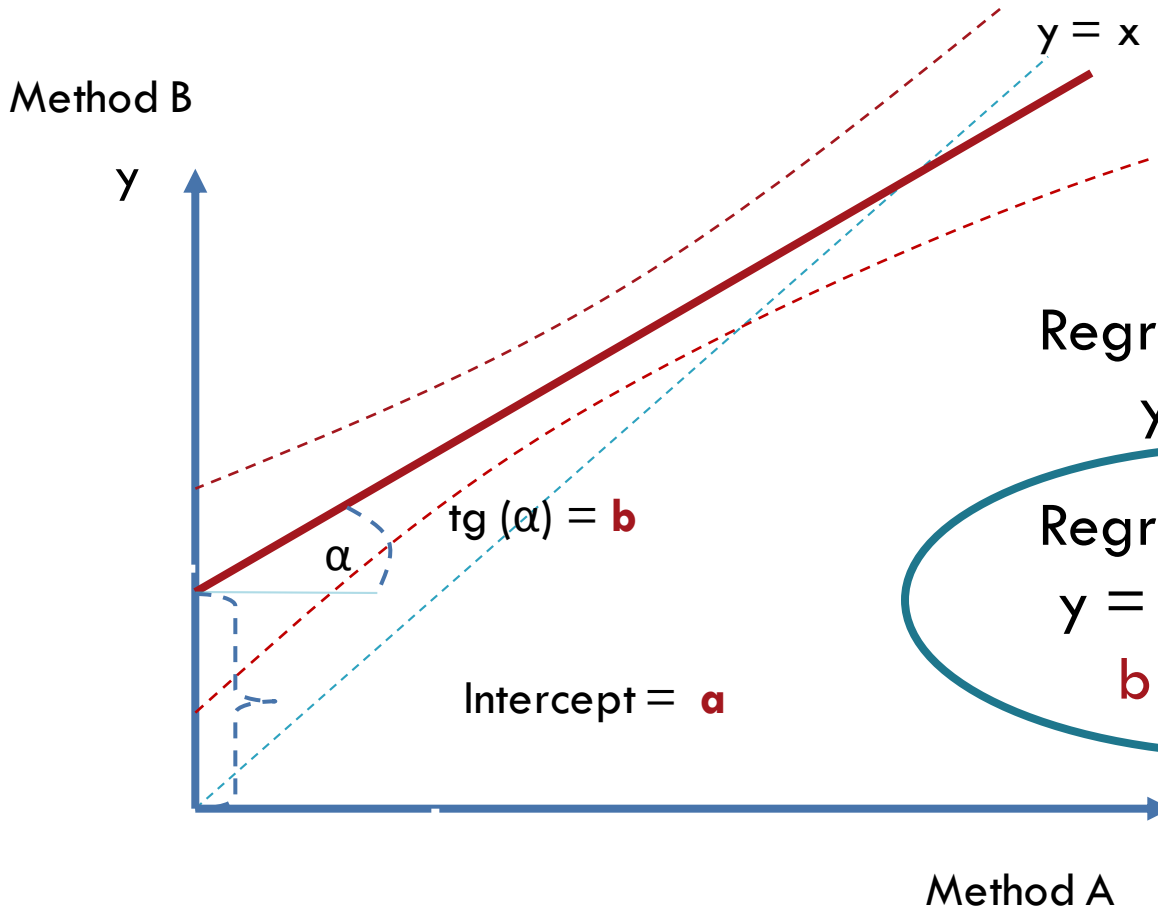
Clinical Department of Laboratory Diagnostics, Clinical Hospital Centre and Department of Medical Informatics, Rijeka University School of Medicine, Rijeka, Croatia

Corresponding author: lidija.bilic-zulle@medri.hr

Bilić-Zulle L. Comparison of methods: Passing and Bablok regression. Biochem Med (Zagreb) 2011;21:49-52.

Linear regression

95% confidence intervals



Regression equation

$y = a + bx$

Regression equation

$y = a\ (95\%\ CI) + b\ (95\%\ CI)\ x$

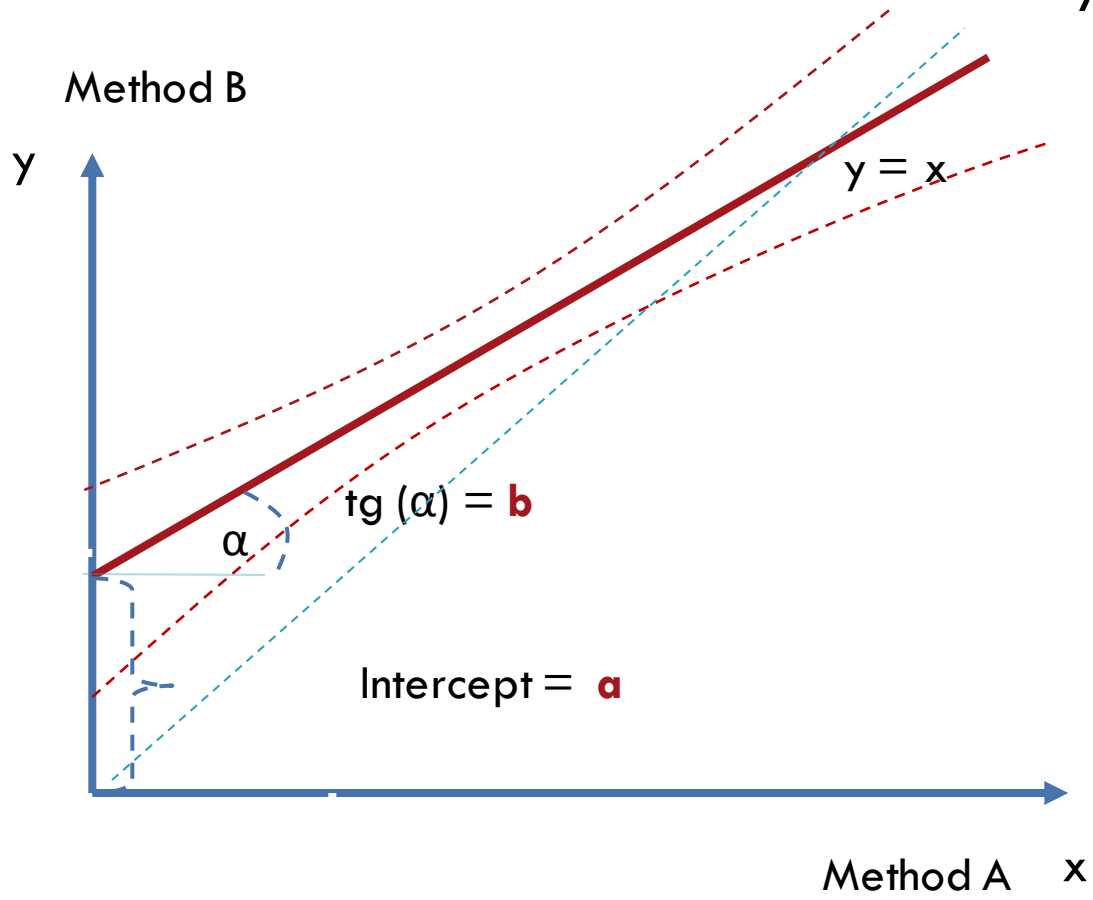
Constant and proportional error

Regression equation

$y = a\ (95\%\ CI) + b\ (95\%\ CI)\ x$

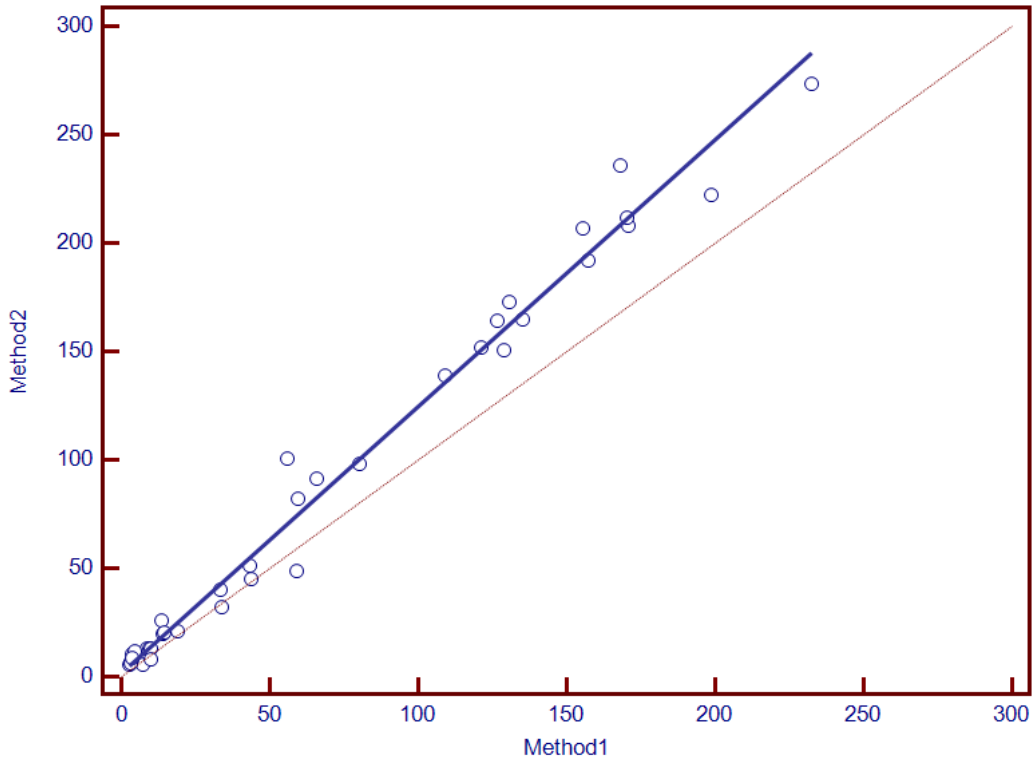
Excluding 0 **Constant error**

Excluding 1 **Proportional error**



Deming regression

- Includes analytical variability of both methods (CV)
- Assumes that errors are independent and normally distributed
- Both methods prone to errors



y = 1.74 (-1.77 to 5.24) + 1.23 (1.16 to 1.30) x

No constant error

Proportional error

Passing-Bablok regression

- Non-parametric method
- No assumptions about distributions of samples
- No assumptions about distributions of errors
- Not sensitive to outliers

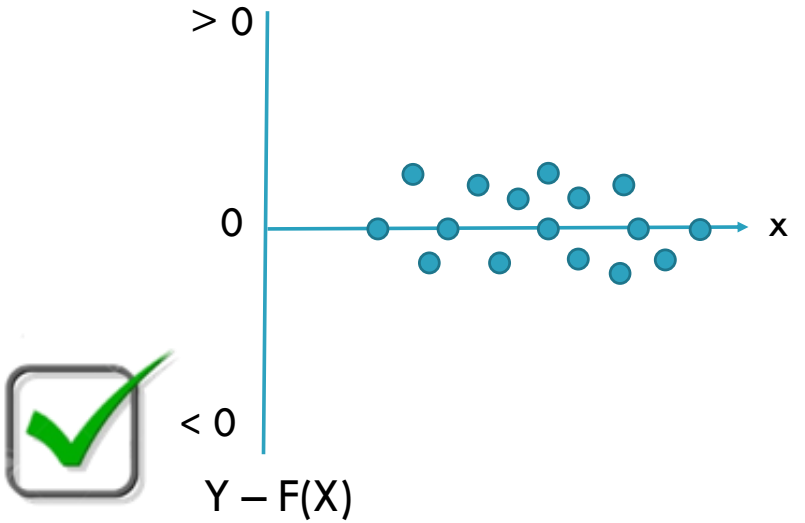
Why don't we recalculate results?

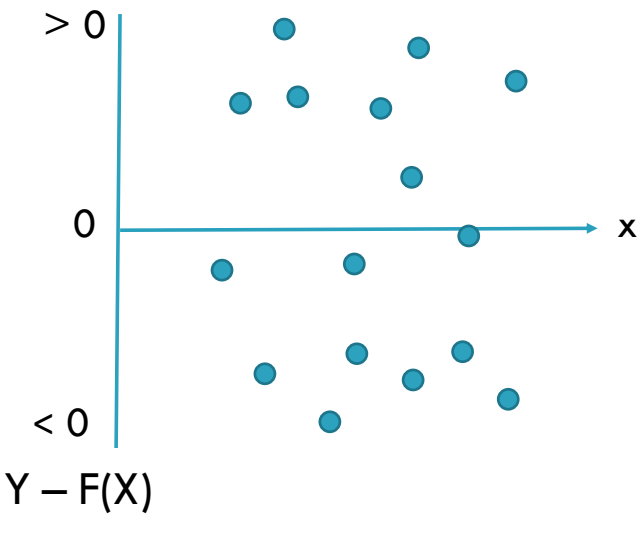
Direct bilirubin (Method 2) =
1.23 x Direct bilirubin (Method 1)

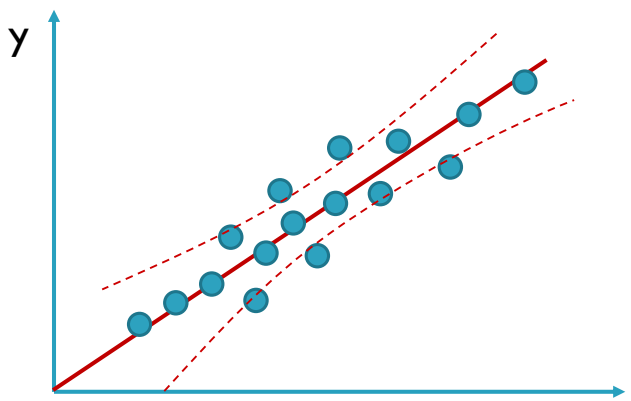
Direct bilirubin (Method 1) =
Direct bilirubin (Method 2) / 1.23

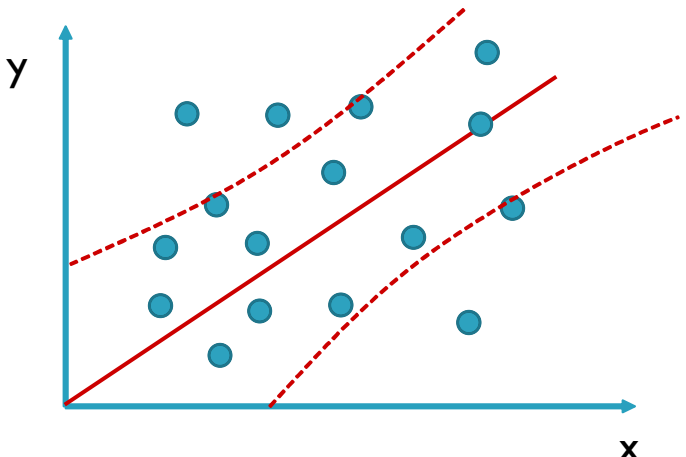
Residual analysis

- How well data fit to the regression model

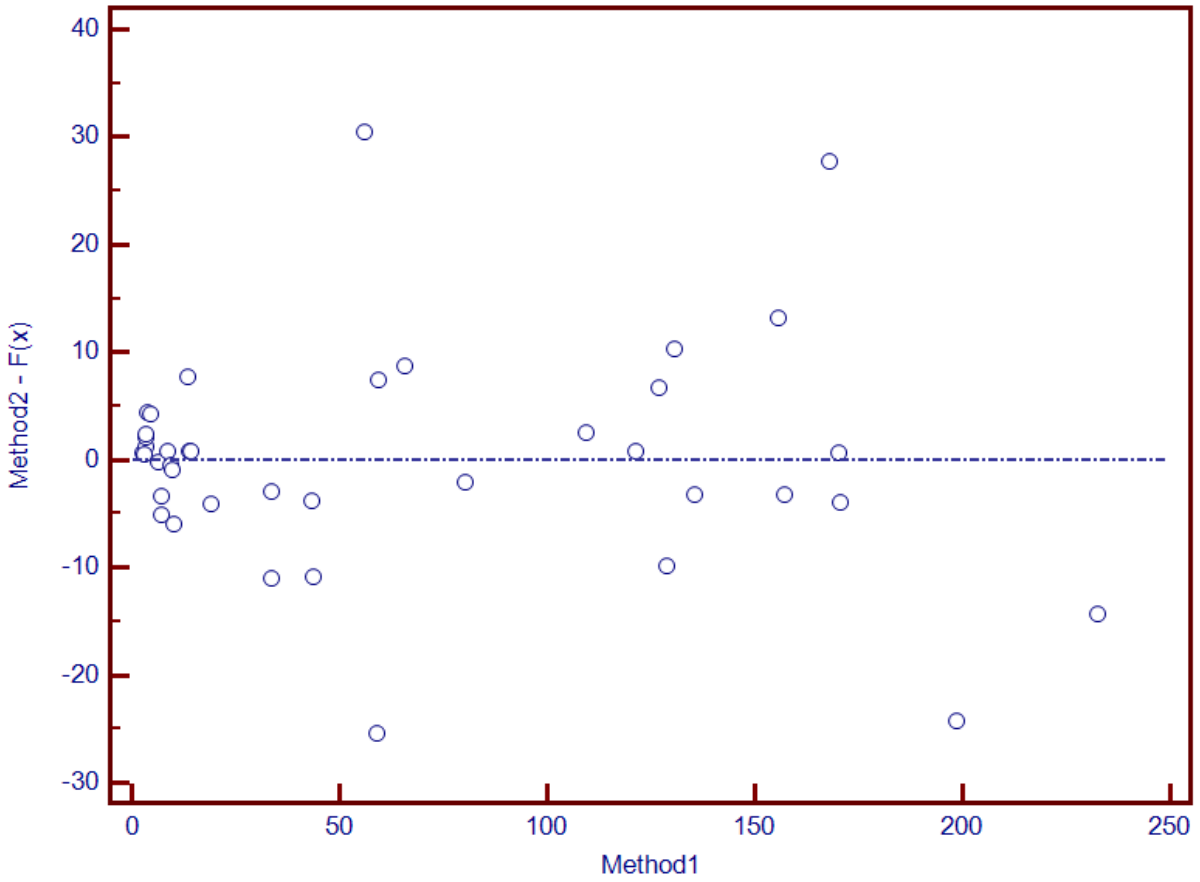








Residual analysis



Differences between measured and calculated values

4.3 Bland-Altman analysis

- Graphical method to compare two measurements technique
- Analyzing differences between measurement pairs

Lessons in biostatistics

Understanding Bland Altman analysis

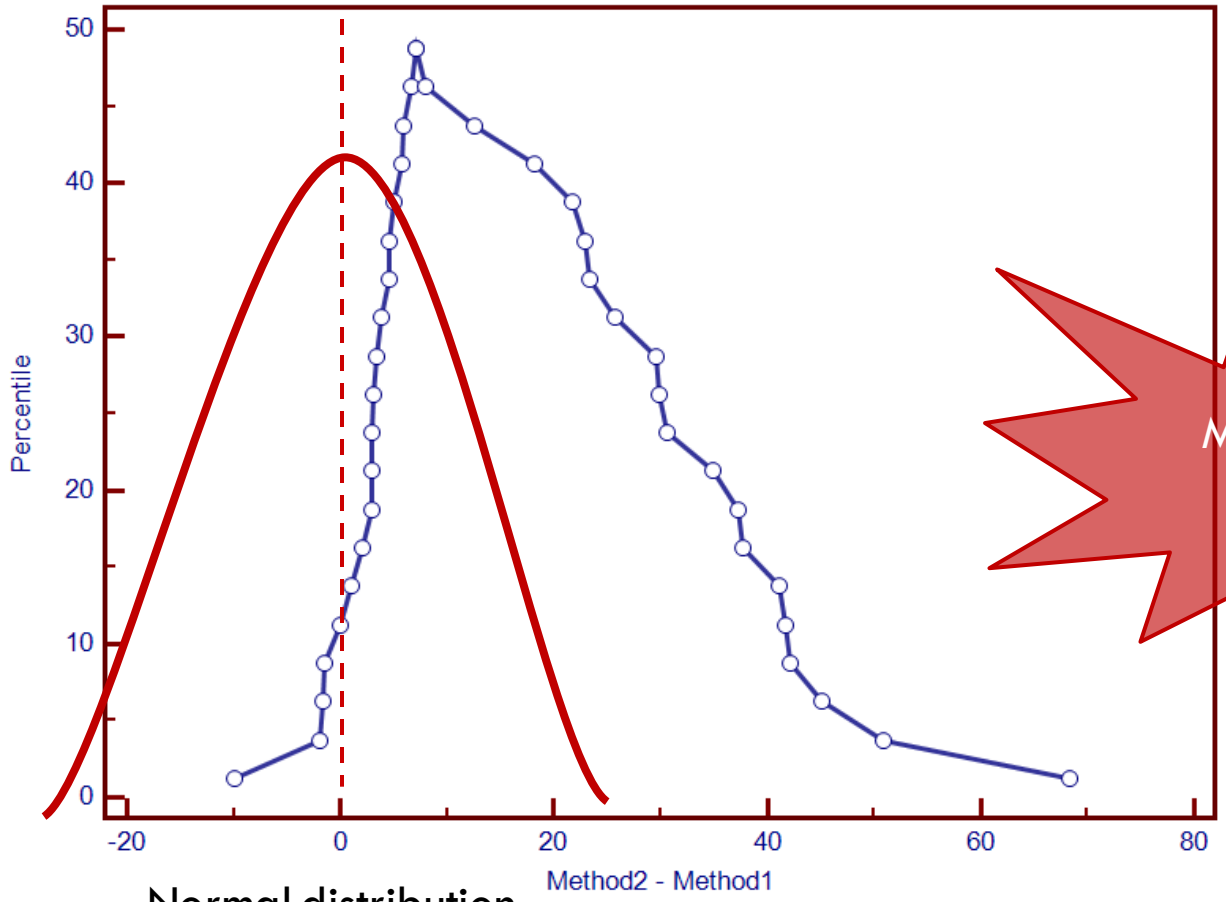
Davide Giavarina

Clinical Chemistry and Hematology Laboratory, San Bortolo Hospital, Vicenza, Italy

Corresponding author: davide.giavarina@ulssvicenza.it

Giavarina D. Understanding Bland Altman analysis. Biochem Med (Zagreb) 2015;25(2):141-51.

Mountain plot

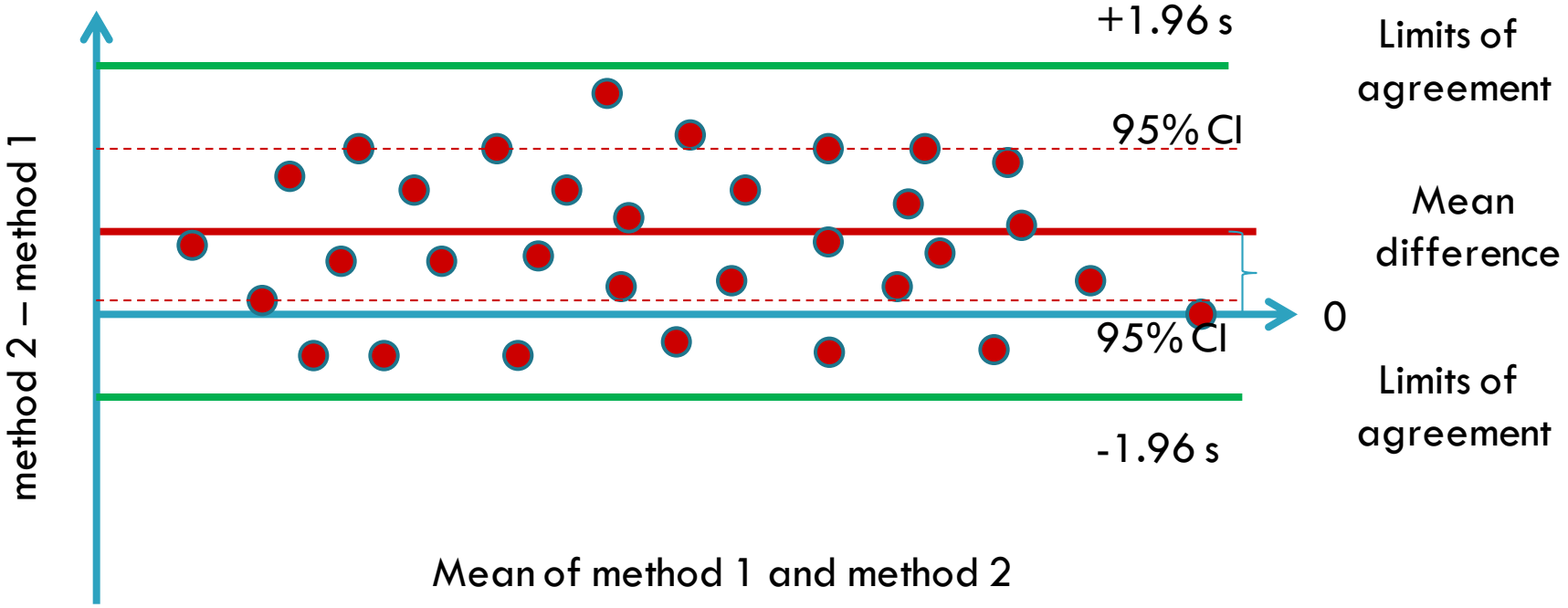


Normal distribution of differences

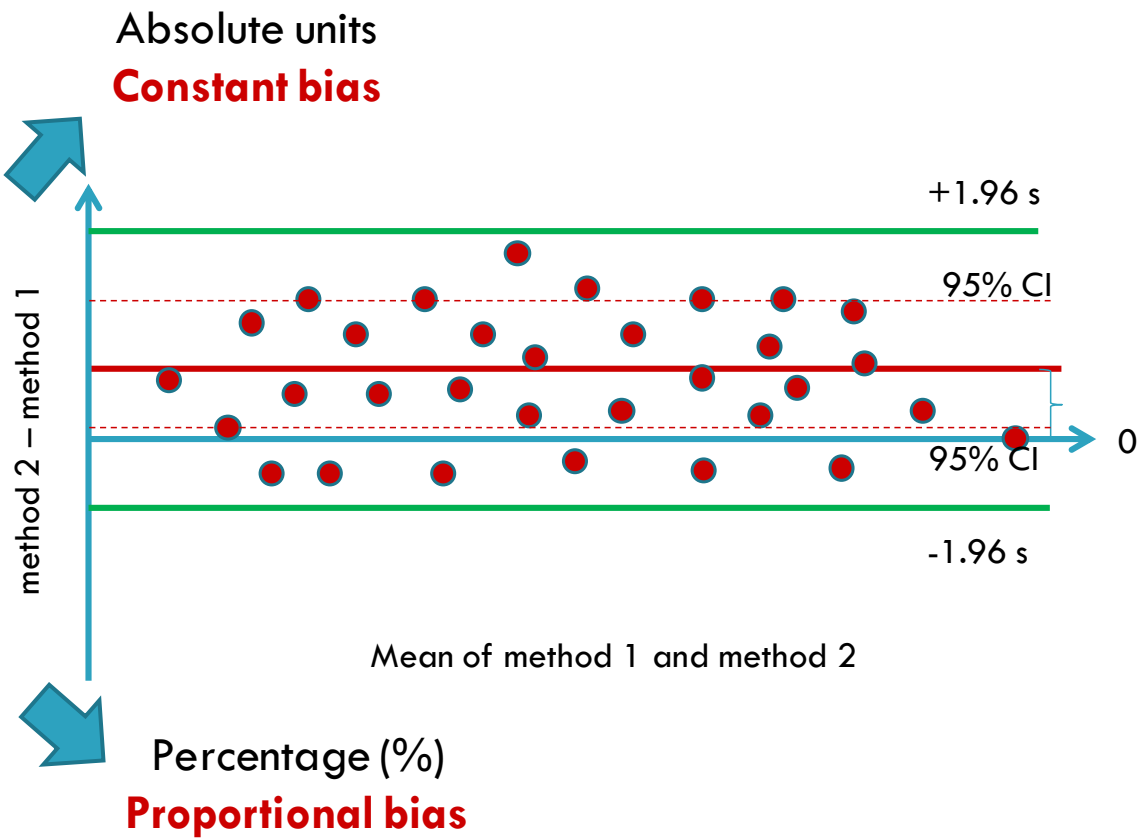
More positive differences

4.3 Bland-Altman analysis

- Plotting differences against:
  - Mean of two methods (no reference method)
  - One method (reference method)



LoA and mean difference



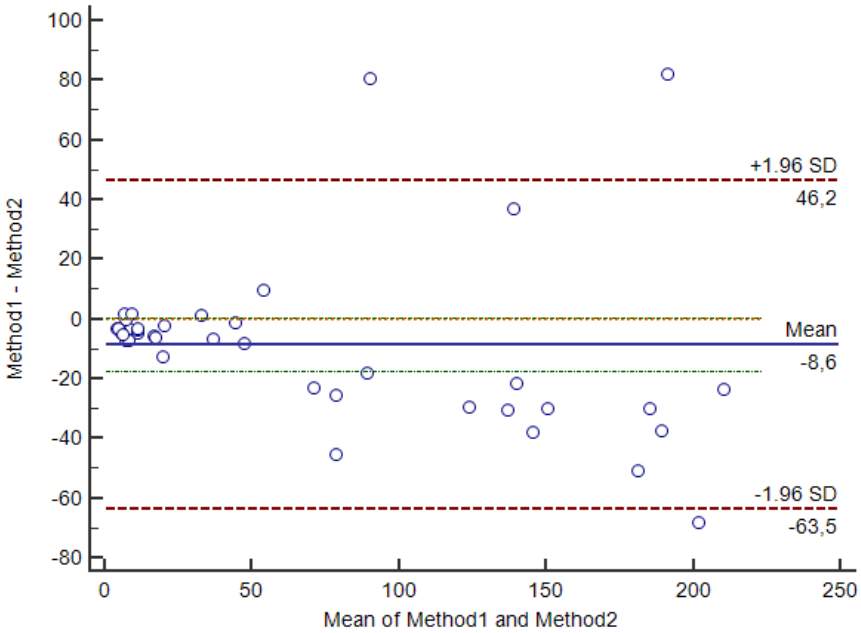
Wide LoA = poor agreement

Including 0 = no constant bias

Excluding 0 = Constant bias

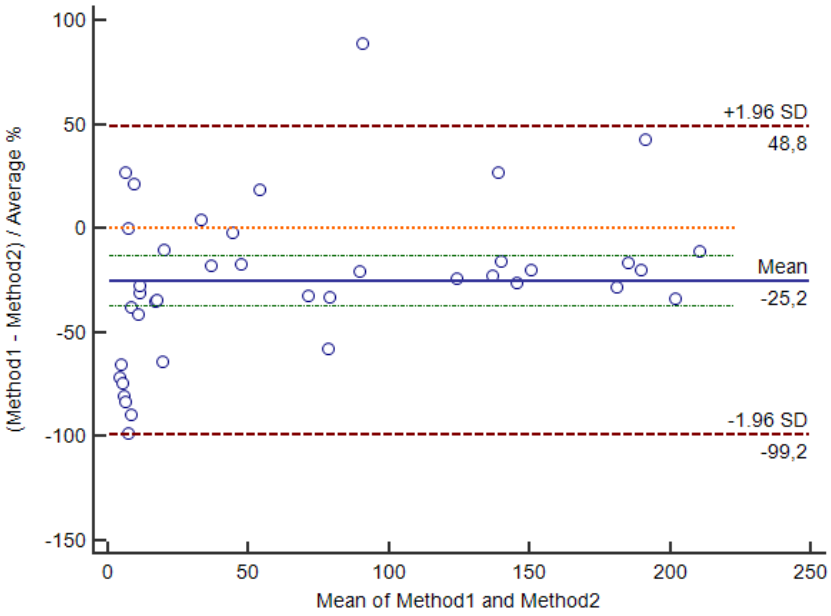
Narrow LoA = good agreement

Bland-Altman analysis



Plotting against mean difference

No constant bias



Plotting against % difference

Proportional bias

5. Data interpretation

Statistical significance ≠ Clinical significance

Comparing values with predefined acceptance criteia

Sunday, 25 Oct, 2015, morning

9:00 – 9:45	Six sigma metrics **Sten Westgard**
9:45 – 10:30	Performance criteria **Gunnar Nordin**
10:30 – 11:15	Biological variation **Sten Westgard**
11:15	Poster award & Closing

Method comparison

- Important laboratory procedure for verification
- Included into validation protocols for new reagents
  - Comparison with the reference method
  - Comparison with different manufacturers
  - Comparison with same manufacturer
- Results are presented in manufacturers declarations

Can we rely on manufacturers declarations?

- Comparing 7 insert sheets for glucose concentration measurement

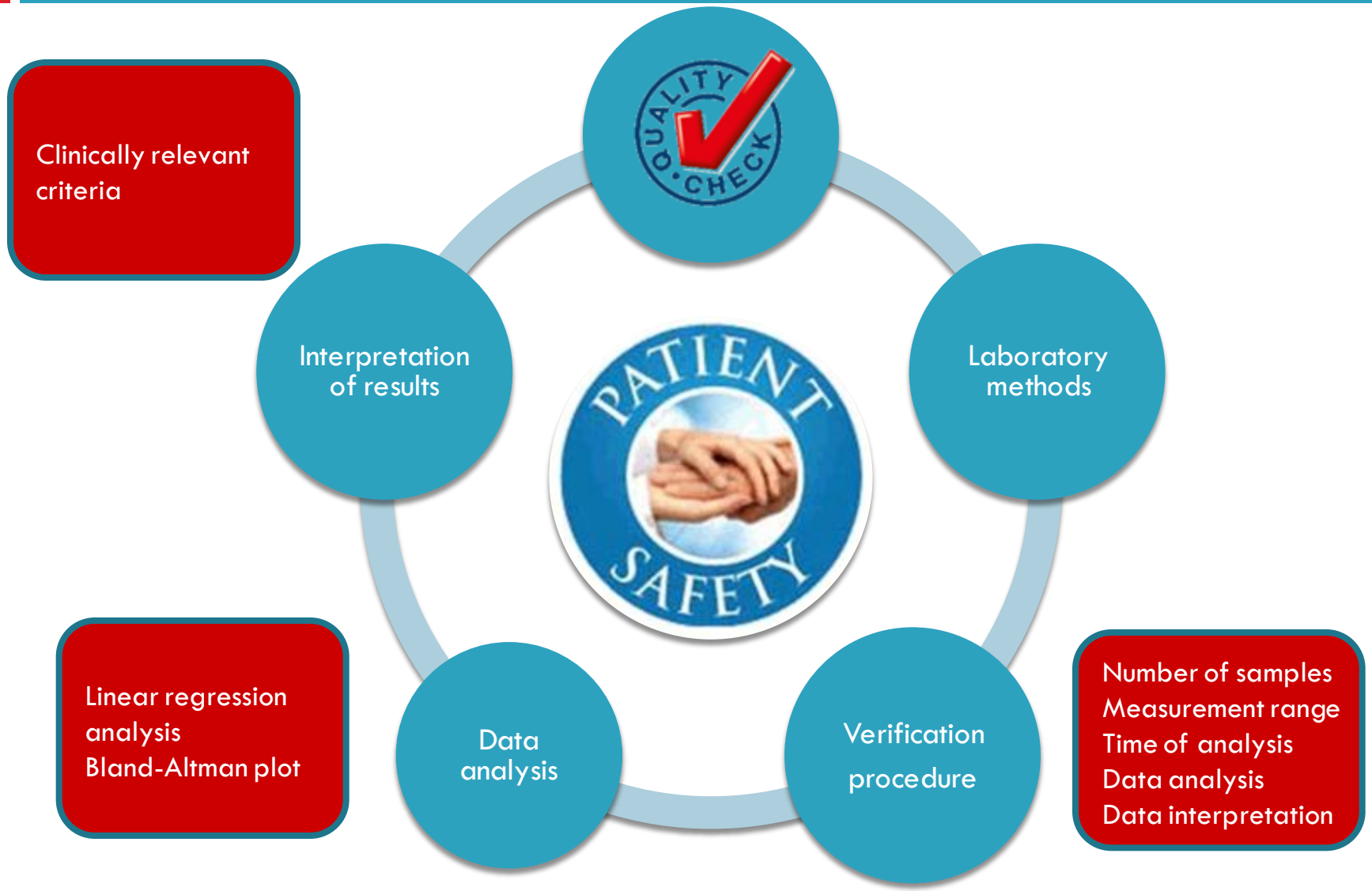
Manufacturer	N	Unit	r	Intercept (95% CI)	Slope (95% CI)
A	102	mg/dL	0.9993	-4.54 (?-?)	1.06 (?-?)
B	117	mmol/L	0.998	-0.081 (?-?)	1.037 (?-?)
C	75	mmol/L	1.000	0.179 (?-?)	0.996 (?-?)
D	43	mg/dL	0.9977	-2.6 (?-?)	1.084 (?-?)
E	?	mg/dL	0.999	0.68 (?-?)	0.99 (?-?)
F	40	mg/dL	0.98	-3.14 (?-?)	0.98 (?-?)
G	60	mmol/L	0.998	0.08 (?-?)	1.008 (?-?)

Correlation for determination of agreement

No BA analysis

No 95% CI for evaluation of bias

To conclude


Clinically relevant criteria
QUALITY CHECK
Laboratory methods
Verification procedure
Data analysis
Interpretation of results
PATIENT SAFETY
Number of samples
Measurement range
Time of analysis
Data analysis
Data interpretation
Linear regression analysis
Bland-Altman plot

Take a home massage

Comparability of methods and analyzers

- Coefficient of correlation doesn't allow conclusions about comparability of methods, but only about linear association between them, even when it is very high (close to 1)
- Regression equation: **Y = 0.67 (-0.15-1.32) + 1.09 (1.03-1.22) x** is an example of proportional bias between methods (95% CI for slope not including 1) without constant bias between methods (95% CI for intercept including 0)
- Regression equation for glucose concentration: **Y = 0.07 (0.01-0.13) + 1.15 (0.85-1.23) x (mmol/L)** is an example of statistically significant, but clinically non-significant constant bias. Value of 0.07 (0.01-0.13) mmol/L glucose is lower than conventional analytical performance of the test