IFCC WG-NB annual meeting 2023

Summary of SFBC WG-NB activities

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Clinical contexts, analytical devices and challenges

About neonatal bilirubinemia

Bilirubin in neonates - Clinical Contexts (1)

In pediatrics, **accurate measurement of total serum bilirubin** (TSB) is of major importance for reliable diagnosis and appropriate management of neonatal jaundice

Clinical indications

- **Screening of neonatal jaundice**
- **Clinical decision of treatment initiation**
  - ✓ therapeutic indication curves (phototherapy and exchange transfusion) based on the total bilirubin (BTS) blood assay
- **Treatment follow-up**
- **Decision to leave** the maternity ward
- **Follow-up after leaving** the maternity ward (HAS recommendation)

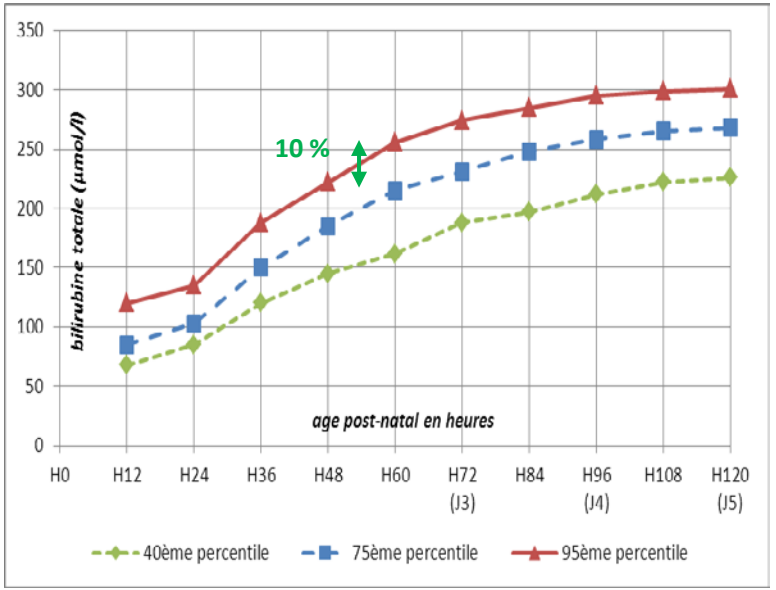
Clinical goal : prevention of severe hyperbilirubinemia

Cortey A et al. Management of jaundice in the newborn ≥ 35 GW: from screening to follow-after discharge. Guidelines for clinical practice – management and treatments after diagnosis. Recommendation SFN 2017 ; 24(2) : 192-203.

Bilirubin in neonates - Clinical Contexts (2)

- As in many countries, the **French Society of Neonatology** (SFN) has issued "Jaundice" recommendations incorporating:
  - ✓ Reference values of bilirubinemia: **nomogram**
  - ✓ Therapeutic indication curves (phototherapy and exchange transfusion) based on the total bilirubin (TSB) blood assay
  - ✓ Keys for the articulation in practice between TcB value (non-invasive quantification) and TSB value (invasive reference quantification) by blood measurement of total bilirubin.
- The **therapeutic indications** for jaundice (phototherapy and exchange transfusion) are based on the **total bilirubin (TSB)** blood assay, interpreted according to
  - ✓ gestational age,
  - ✓ postnatal age in hours and
  - ✓ conditions of vulnerability to bilirubin toxicity

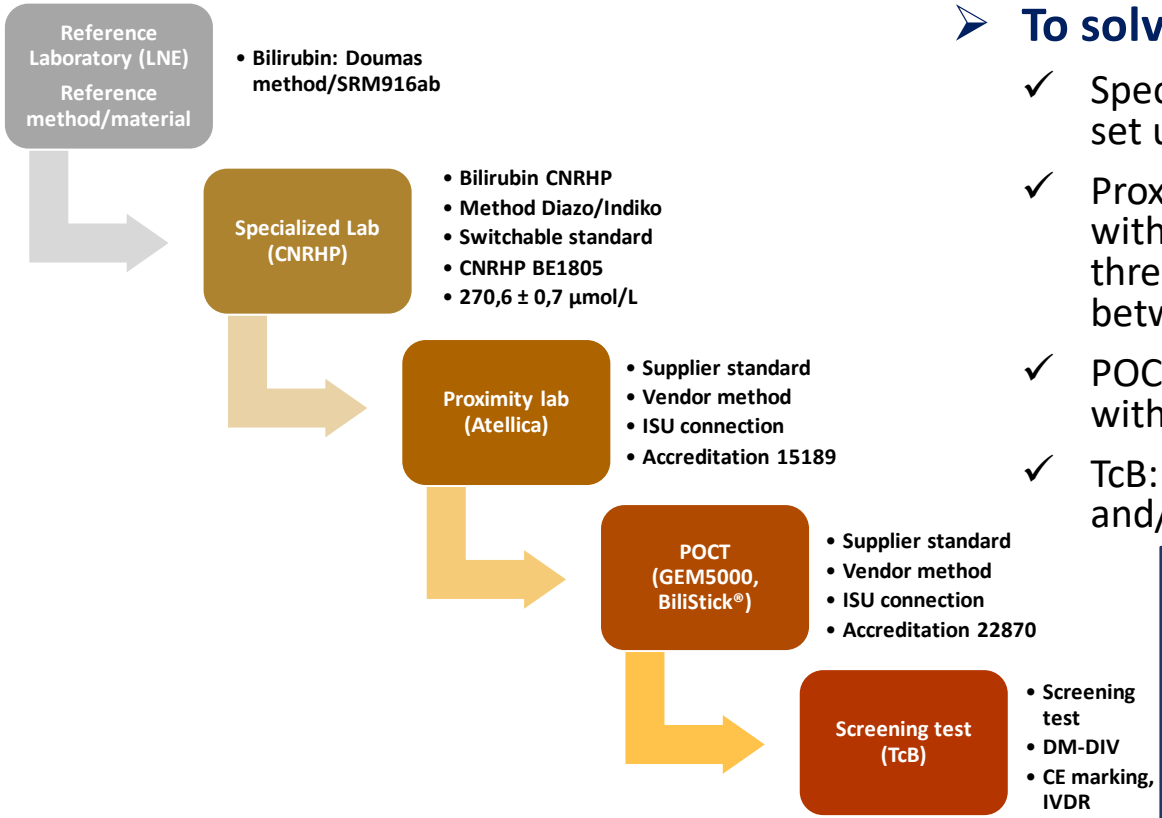
BTS if TcB >75^{e}percentile or >250 µmol/L
Emergency if blinking or > 957 mep.



Bhutani VK, Johnson L: A proposal to prevent severe neonatal hyperbilirubinemia and kernicterus. J Perinatol. 2009 Feb;29 Suppl 1:S61-7
Cortey A.,Tourneux P., Bedu A., Renesme L., Raignoux J., Casper C., Truffert P. Management of jaundice in the newborn ≥ 35 GW: from screening to follow-after discharge Guidelines for clinical practice – management and treatments after diagnosis. Recommendation SFN 2015
American Academy of Pediatrics, Subcommittee on Hyperbilirubinemia. Management of hyperbilirubinemia in the newborn infant 35 or more weeks of gestation [published correction appears in Pediatrics. 2004;114:1138]. Pediatrics. 2004;114 :297 –316

Analytical goal: provide a reliable and accurate result (<10 % total error), even in POCT, for an optimized clinical decision

Laboratory needs and analytical challenges



➢ To solve at each level

- ✓ Specialized labs: work on IS connection, set up national harmonization studies
- ✓ Proximity lab: select techniques, assist with interpretation against consensual thresholds and monitor comparability between laboratory (EQA/PT)
- ✓ POCT: same + comparability monitoring with the central lab
- ✓ TcB: help in choosing devices + lab and/or POCT results comparability

In response to these challenges, the CNRHP lab has carried out several local or national studies to progress towards better emergency interpretation of neonatal bilirubin results.

CNRHP Study
Connection to International System of Units

STANDARDIZATION OF SERUM TOTAL BILIRUBIN MEASUREMENT FOR IMPROVED DIAGNOSIS AND MANAGEMENT OF NEONATAL JAUNDICE

Euromedlab – Athènes – Juin 2017

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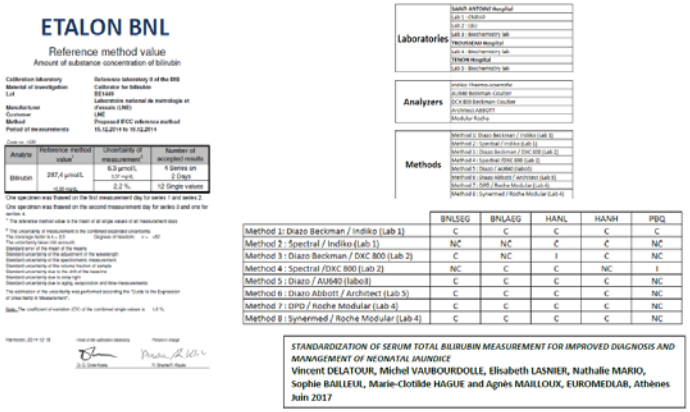
Identification d'un calibrant commutable pour réaliser une calibration *in silico*



- **This study allowed to**
  - ✓ **Check the commutability** of a CNRHP BNL standard and an LNE HAN control material for 8 automaton/technical pairs (exception spectral method)
  - ✓ **Assign a value** to the CNRHP BNL standard by connection carried out by the LNE
- **And to be able to use**
  - ✓ **The CNRHP-BNL standard** for linking the total bilirubin assays carried out at the CNRHP and thus determining the expected values of Bilirubin during a harmonization study
  - ✓ **The switchable control samples** with assigned values

Objectives of the 2015 study

Produce candidate CRMs that could be used to improve harmonization of bilirubin assays (calibration materials) and monitor it (trueness verifiers)

- BNL standard prepared by CNRHP
- 2 pools of human frozen serum : 1 pool with bilirubin conc. between 15 and 30 µM (Pool « HANL ») and one pool between 220 and 250 µM (Pool « HANH »)
- Target values assigned with Hanover's candidate RMP
- Materials commutability evaluated. Commutability of an EQA material also evaluated
- *In silico* recalibration to evaluate the impact of recalibrating assays

Samples production and characteristics

- HANL & HANH pools produced from a sufficient number of clinical samples with very low (HANL pool) or very high (HANH pool) bilirubin concentrations.
- Single donor samples measured using a routine method and stored at -80°C until the desired volume is available to prepare the pools.
- BNL 1449 standard produced on 02/12/2014 at CNRHP from material issued by the French blood center.
- 2 batches of the BNL 1449 standard produced: with (BNLAEG) and without (BNLSEG) addition of ethylene glycol in order to evaluate the impact of the addition of this cryopreservative on the commutability of the BNL standard.
- 3 pools of frozen serum produced in Hannover for quality control purposes (QC1, QC2, QC3).
- PBQ sample: freeze-dried serum pool Probioqual (EQA material)

Assignation de valeurs de référence

- For samples HANL and HANH, BNLAEG, QC1, QC2 and QC3, ref values assigned with the IFCC candidate reference method
- Method developed in collaboration with Roche Diagnostics
- Similar to the Doumas method no calibration: total bilirubin concentration determined spectrophotometrically with a unique molar extinction coefficient for the different forms of bilirubin.
- measurements conducted in the reference laboratory of RfB in Hannover
- Method validated via several inter-comparisons: results equivalent to those provided by the original Doumas method:
  - bias less than 1.7% compared to the consensus value
  - CV around 1% (n=12, 1 triplicate performed on 4 different days)
  - relative expanded uncertainty was about 2.2%

Samples

- 50 x1 mL of « HANL» at 25,9 ± 0,6 µmol/L
- 50 x 1 mL du pool « HANH » at 254,9 ± 5,6 µmol/L
- 3 x 1 mL of « QC1 » at 21,7 ± 0,6 µmol/L
- 3 x 1 mL of « QC2 » at 69,7 ± 1,8 µmol/L
- 3 x 1 mL of « QC3 » at 156,8 ± 4,0 µmol/L

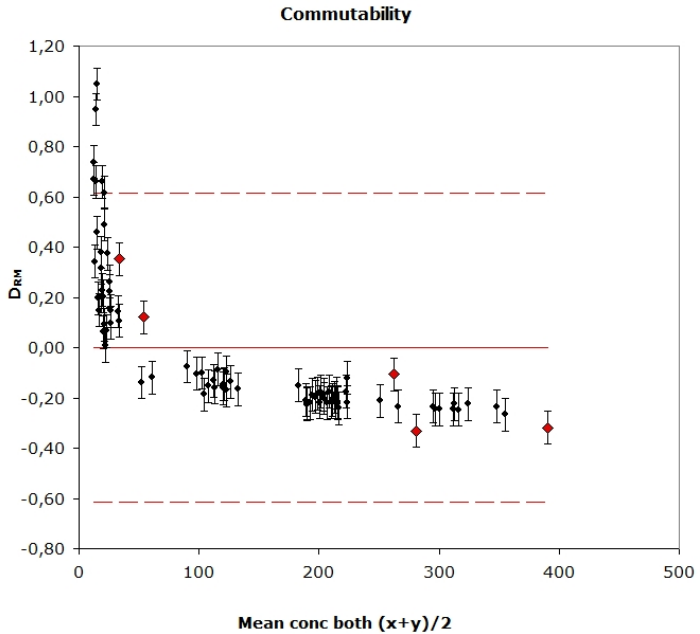
Clinical specimens

- In principle, native samples = single donations in the form of fresh unpooled unfrozen serum
- Difficulty in obtaining a sufficient number of fresh non-pooled samples
  → frozen samples
- 83 non-pooled frozen samples recruited by CNRHP with bilirubin concentrations ranging from approximately 6 to 400 µM.
- To ensure that the frozen clinical samples are commutable to fresh samples,
  5 pools of fresh serum were prepared from newborns sera to cover the high values (BBFP) and 5 unpooled fresh samples from adult donors were recruited to cover the low values (AFNP).

Nomenclature			
Code	Description	Matrice	Nb ech
PS	Patient sample with Low concentration	pool congelé	25
PS	Patient sample with Medium concentration	pool congelé	23
PS	Patient sample with High concentration	pool congelé	25
BBFP	Patient sample - Pool Bébé Frais	Pool frais bébés	5
AFNP	Patient sample - Adult Frais non poolé	Single donation Fraiche non poolée	5
BNLSEG	Etalon BNL sans Ethylene Glycol	Single donation liquide congelée	1
BNLAEG	Etalon BNL avec Ethylene Glycol	Single donation liquide congelée + EG	1
HANL	Hanovre Pool Low concentration	pool congelé	1
HANH	Hanovre Pool High concentration	pool congelé	1
PBQ	ProBioQual	pool lyophilisé	1

Commutability evaluation: statistical analysis

- The statistical analysis was first performed in full scale
- Commutability criterion C too high: 41.8% (SD=21.8%, n=28).
- Leads to the wrong conclusion that all samples are commutable



Statistical analysis commutability

- Statistical analysis split in 2 concentration ranges: > and < 50µM.
- Low" concentrations = less than 50 µM
- Medium" concentrations = between 50 and 150 µM
- High" concentrations = above 150 µM
- Samples whose commutability has been evaluated either very low (HANL, PBQ) or very high (BNLSEG, BNLAEG, HANH) → samples with average concentrations (between 60 and 150 µM) finally not used
- This situation had been anticipated with 3 analytical sequences:
  - 1 dedicated to low values (25 frozen PS + 5 fresh non-pooled PS from adult donors (AFNP) + HANL + PBQ
  - 1 for intermediate values (not used)
  - 1 for high values (32 frozen PS + 5 fresh pools prepared from baby sera (BBFP) + BNLSEG + BNLAEG + HANH

Data analysis

- With the split of the statistical analysis into 2 concentration zones, much more reasonable acceptability criteria for high concentrations: C=10.5% (SD=6.3%, n=28) but for low concentrations, very high statistical acceptability criterion , which is inappropriate for decision making because too little demanding: C= 42.7% (SD=23.3%, n=28).
- Situation due to very heterogeneous behavior of samples with low bilirubin concentration.
- Not due to the design of the statistical study but rather to the performance of the assay methods (difference in specificity of the different methods or strong impact of interferences)
- Use of a much more stringent fixed acceptability criterion close to the acceptability criterion for high concentrations: C=10%.
- Very high number of clinical samples found to be non-commutable
- Justified by the fact that the samples evaluated are intended to be used as standards

Data analysis

BNLAEG	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8
Methode 1	-	NC	I	C	C	C	C	C
Methode 2	-	-	NC	I	NC	NC	NC	NC
Methode 3	-	-	-	NC	I	I	I	I
Methode 4	-	-	-	-	C	C	C	C
Methode 5	-	-	-	-	-	C	C	C
Methode 6	-	-	-	-	-	-	C	C
Methode 7	-	-	-	-	-	-	-	C
Methode 8	-	-	-	-	-	-	-	-

BNLAEG	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8	Moyenne	SD
Methode 1		12,0%	20,3%	3,3%	1,0%	0,9%	1,4%	0,2%	5,6%	7,7%
Methode 2			32,4%	15,3%	13,0%	11,2%	10,6%	10,8%	15,8%	8,3%
Methode 3				17,0%	19,3%	21,2%	21,7%	21,5%	21,9%	4,9%
Methode 4					2,3%	4,2%	4,7%	4,5%	7,3%	6,1%
Methode 5						1,8%	2,4%	2,2%	6,0%	7,2%
Methode 6							0,5%	0,4%	5,7%	7,8%
Methode 7								0,2%	5,9%	7,8%
Methode 8									5,7%	8,0%

Hanover Pool at low concentration (HANL)

HANL	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8
Methode 1	-	I	C	I	C	C	C	C
Methode 2	-	-	C	I	I	C	I	I
Methode 3	-	-	-	I	I	I	I	I
Methode 4	-	-	-	-	I	I	I	I
Methode 5	-	-	-	-	-	C	C	C
Methode 6	-	-	-	-	-	-	C	C
Methode 7	-	-	-	-	-	-	-	C
Methode 8	-	-	-	-	-	-	-	-

HANL	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8	Moyenne	SD
Methode 1		1,8%	9,0%	2,2%	2,2%	6,7%	7,2%	3,7%	4,7%	2,9%
Methode 2			4,9%	5,7%	0,6%	2,9%	3,2%	0,0%	3,2%	1,9%
Methode 3				13,8%	6,9%	11,5%	11,9%	8,3%	9,5%	3,1%
Methode 4					6,5%	3,0%	2,7%	5,9%	5,7%	4,0%
Methode 5						3,8%	4,2%	0,9%	3,6%	2,5%
Methode 6							4,2%	3,6%	5,1%	3,1%
Methode 7								4,1%	5,3%	3,2%
Methode 8									3,8%	2,8%

Pool Hanover Haute concentration (HANH)

HANH	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8
Methode 1	-	NC	C	C	NC	NC	NC	C
Methode 2	-	-	C	C	C	C	C	C
Methode 3	-	-	-	C	C	C	C	C
Methode 4	-	-	-	-	I	C	C	C
Methode 5	-	-	-	-	-	C	C	C
Methode 6	-	-	-	-	-	-	C	C
Methode 7	-	-	-	-	-	-	-	I
Methode 8	-	-	-	-	-	-	-	-

HANH	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8	Moyenne	SD
Methode 1		10,6%	4,3%	2,6%	12,6%	7,9%	9,7%	1,1%	7,0%	4,4%
Methode 2			6,2%	13,2%	2,1%	2,6%	0,9%	0,2%	5,9%	5,0%
Methode 3				7,0%	8,3%	3,6%	5,3%	6,4%	5,9%	1,6%
Methode 4					15,3%	10,6%	12,3%	13,4%	10,6%	4,4%
Methode 5						4,7%	3,0%	1,9%	6,8%	5,4%
Methode 6							1,7%	2,8%	4,8%	3,2%
Methode 7								1,1%	4,8%	4,5%
Methode 8									3,8%	4,7%

BNL standard without ethylene glycol (BNLSEG)

BNLSEG	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8
Methode 1	-	NC	C	C	C	C	C	C
Methode 2	-	-	C	C	I	NC	NC	NC
Methode 3	-	-	-	NC	C	C	C	C
Methode 4	-	-	-	-	C	I	I	C
Methode 5	-	-	-	-	-	C	I	I
Methode 6	-	-	-	-	-	-	C	C
Methode 7	-	-	-	-	-	-	-	C
Methode 8	-	-	-	-	-	-	-	-

BNLSEG	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8	Moyenne	SD
Methode 1		10,8%	3,8%	11,5%	3,0%	3,3%	2,1%	0,1%	4,9%	4,4%
Methode 2			14,6%	0,6%	7,8%	14,1%	13,0%	12,8%	10,2%	5,3%
Methode 3				15,3%	6,8%	0,5%	1,7%	1,8%	6,4%	6,2%
Methode 4					8,4%	14,7%	13,6%	13,5%	11,1%	5,2%
Methode 5						6,3%	5,1%	5,0%	6,1%	1,8%
Methode 6							1,2%	1,3%	5,9%	6,1%
Methode 7								0,1%	5,2%	5,7%
Methode 8									4,9%	5,9%

BNL standard with ethylene glycol (BNLAEG)

BNLAEG	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8
Methode 1	-	NC	I	C	C	C	C	C
Methode 2	-	-	NC	I	NC	NC	NC	NC
Methode 3	-	-	-	NC	I	I	I	I
Methode 4	-	-	-	-	C	C	C	C
Methode 5	-	-	-	-	-	C	C	C
Methode 6	-	-	-	-	-	-	C	C
Methode 7	-	-	-	-	-	-	-	C
Methode 8	-	-	-	-	-	-	-	-

BNLAEG	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8	Moyenne	SD
Methode 1		12,0%	20,3%	3,3%	1,0%	0,9%	1,4%	0,2%	5,6%	7,7%
Methode 2			32,4%	15,3%	13,0%	11,2%	10,6%	10,8%	15,8%	8,3%
Methode 3				17,0%	19,3%	21,2%	21,7%	21,5%	21,9%	4,9%
Methode 4					2,3%	4,2%	4,7%	4,5%	7,3%	6,1%
Methode 5						1,8%	2,4%	2,2%	6,0%	7,2%
Methode 6							0,5%	0,4%	5,7%	7,8%
Methode 7								0,2%	5,9%	7,8%
Methode 8									5,7%	8,0%

Effet of PEG

BNLSEG	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8	Moyenne	SD
Methode 1		10,8%	3,8%	11,5%	3,0%	3,3%	2,1%	0,1%	4,9%	4,4%
Methode 2			14,6%	0,6%	7,8%	14,1%	13,0%	12,8%	10,2%	5,3%
Methode 3				15,3%	6,8%	0,5%	1,7%	1,8%	6,4%	6,2%
Methode 4					8,4%	14,7%	13,6%	13,5%	11,1%	5,2%
Methode 5						6,3%	5,1%	5,0%	6,1%	1,8%
Methode 6							1,2%	1,3%	5,9%	6,1%
Methode 7								0,1%	5,2%	5,7%
Methode 8									4,9%	5,9%

BNLAEG	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8	Moyenne	SD
Methode 1		12,0%	20,3%	3,3%	1,0%	0,9%	1,4%	0,2%	5,6%	7,7%
Methode 2			32,4%	15,3%	13,0%	11,2%	10,6%	10,8%	15,8%	8,3%
Methode 3				17,0%	19,3%	21,2%	21,7%	21,5%	21,9%	4,9%
Methode 4					2,3%	4,2%	4,7%	4,5%	7,3%	6,1%
Methode 5						1,8%	2,4%	2,2%	6,0%	7,2%
Methode 6							0,5%	0,4%	5,7%	7,8%
Methode 7								0,2%	5,9%	7,8%
Methode 8									5,7%	8,0%

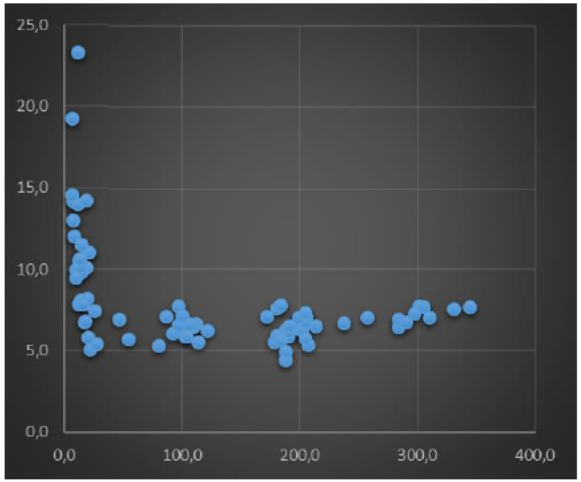
ProBioQual (PBQ)

PBQ	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8
Methode 1	-	I	I	I	I	C	C	C
Methode 2	-	-	I	C	I	NC	NC	NC
Methode 3	-	-	-	I	I	I	I	I
Methode 4	-	-	-	-	I	I	I	I
Methode 5	-	-	-	-	-	NC	NC	NC
Methode 6	-	-	-	-	-	-	NC	I
Methode 7	-	-	-	-	-	-	-	I
Methode 8	-	-	-	-	-	-	-	-

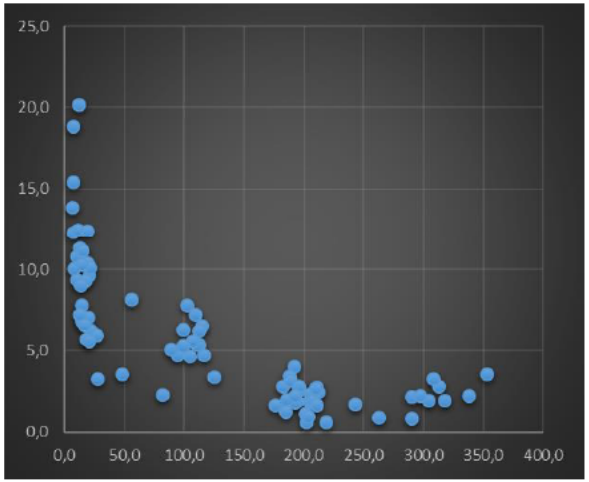
PBQ	Methode 1	Methode 2	Methode 3	Methode 4	Methode 5	Methode 6	Methode 7	Methode 8	Moyenne	SD
Methode 1		20,0%	14,1%	2,1%	8,7%	1,1%	0,8%	1,4%	6,9%	7,6%
Methode 2			6,0%	1,2%	14,9%	30,4%	28,7%	28,2%	16,9%	11,8%
Methode 3				15,0%	8,3%	15,9%	16,2%	14,8%	12,9%	4,0%
Methode 4					3,1%	12,4%	10,7%	10,2%	7,8%	5,6%
Methode 5						19,0%	17,4%	16,9%	12,6%	5,9%
Methode 6							17,4%	9,3%	15,1%	9,0%
Methode 7								9,0%	14,3%	8,4%
Methode 8									12,8%	6,5%

In silico recalibration

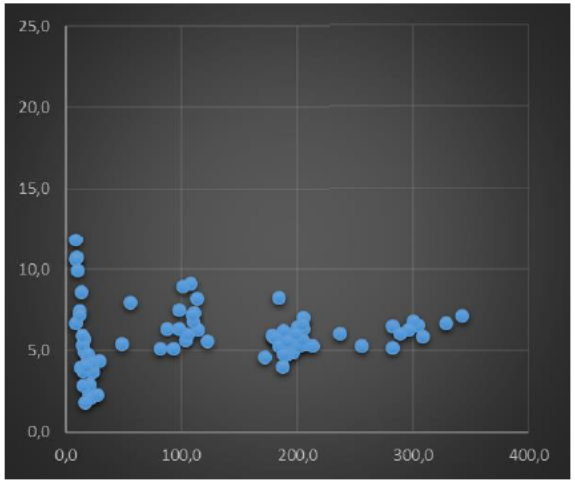
Inter-laboratory variation – CV% = f(concentration - µmol/L)



Raw data
Mean CV = 5,5%



1-point recalibration
Mean CV = 2,0%



2-point recalibration
Mean CV = 5,0%

First Study Conclusions (1)

- Hanover Pools HANL and HANH + BNL standards with and without ethylene glycol are commutable for most but not all methods.
- BNL standard with ethylene glycol not suitable for calibration of methods 2 and 3 nor for evaluation of their accuracy. The addition of ethylene glycol degrades the commutability of the BNL standard for methods 2 and 3.
- HANL pool suitable to calibrate all methods (except method 3?)
- Due to the high variability observed at low bilirubin values, it is unlikely that using this sample as a standard will improve the accuracy of results in this concentration range, but better comparability of results is expected.
- Pool HANH suitable for calibration of all methods except perhaps method 4. Pool HANH superior to BNL standard without ethylene glycol, which is itself more suitable than BNL standard with ethylene glycol for use as a standard or for accuracy verification purposes.
- Very low commutability of the ProBioQual freeze-dried sample

First Study Conclusions (2)

	BNLSEG	BNLAEG	HANL	HANH	PBQ
méthode 1: diazo Beckman /indiko (labo1)	C	C	C	C	C
méthode 2 : Spectrale/ indiko (labo1)	NC	NC	C	C	NC
méthode 3 : diazo Beckman /DXC 800 (labo2)	C	NC	I	C	NC
méthode 4 : Spectrale /DXC 800 (labo2)	NC	C	C	NC	I
méthode 5 : Diazo/AU640 (labo3)	C	C	C	C	NC
méthode 6 : diazo Abbott/ Architect (labo5)	C	C	C	C	NC
méthode 7 : DPD/Modular (labo4)	C	C	C	C	NC
méthode 8 : Synermed/Modular (labo4)	C	C	C	C	NC

- Commutability of BNL standards and Hanover pools sufficient to consider their use for calibrating most methods
- Highly heterogeneous behavior of the different methods for low bilirubin values, it is not reasonable to perform a 2-point calibration with one point below 50µM and one point above, as all intermediate values would not be covered.

French National multicenter study CNRHP – SFBC – CNBH
Harmonization between French labs

Synthèse

ANNALES DE BIOLOGIE CLINIQUE

Ann Biol Clin 2020 ; 78 (4) : 383-97

Recommandations analytiques et cliniques pour l'utilisation de la bilirubinémie en néonatalogie

Analytical and clinical guidelines on neonatal bilirubinemia

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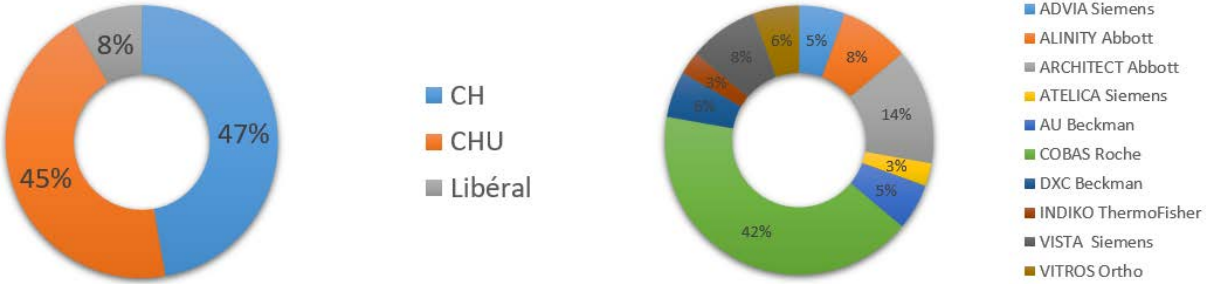
SFBC

We then conducted a national harmonization study with the SFBC. Thirty-six laboratories participated in this study with most of the analyzers used in France but an over-representation of ROCHE analyzers.



Laboratories repartition (n=36)

We had a representative sample of blood bilirubin measurement methods in France

French National multicenter study CNRHP – SFBC – CNBH
Harmonization between French labs

METHODS

36 French clinical laboratories were included in the study, using the 10 most popular routine assays. Each lab analyzed in a single run 30 pools prepared from adults and neonates leftovers, our secondary standard and 2 trueness verifiers in triplicate. Expected values were determined in CNRHP lab using secondary standard with reference method assigned value. Results were further analyzed in 2 sub-groups: TSB<150 µM and TSB >150 µM. Following analytical acceptance limits were proposed at TSB >150 µM :for imprecision estimation maximum CV 3.2 % (repeatability), 4.2 % (interlaboratory variation - same technique), 10 % (all techniques) and for accuracy approach 10 % limit bias. These limits take into accounts the clinical guidelines for diagnosis and therapeutic monitoring of neonatal jaundice.

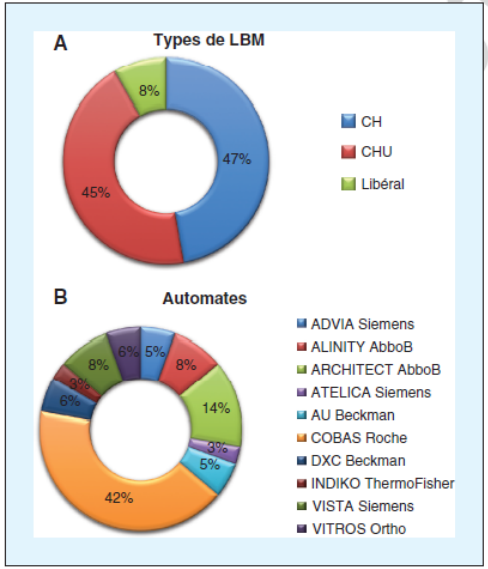
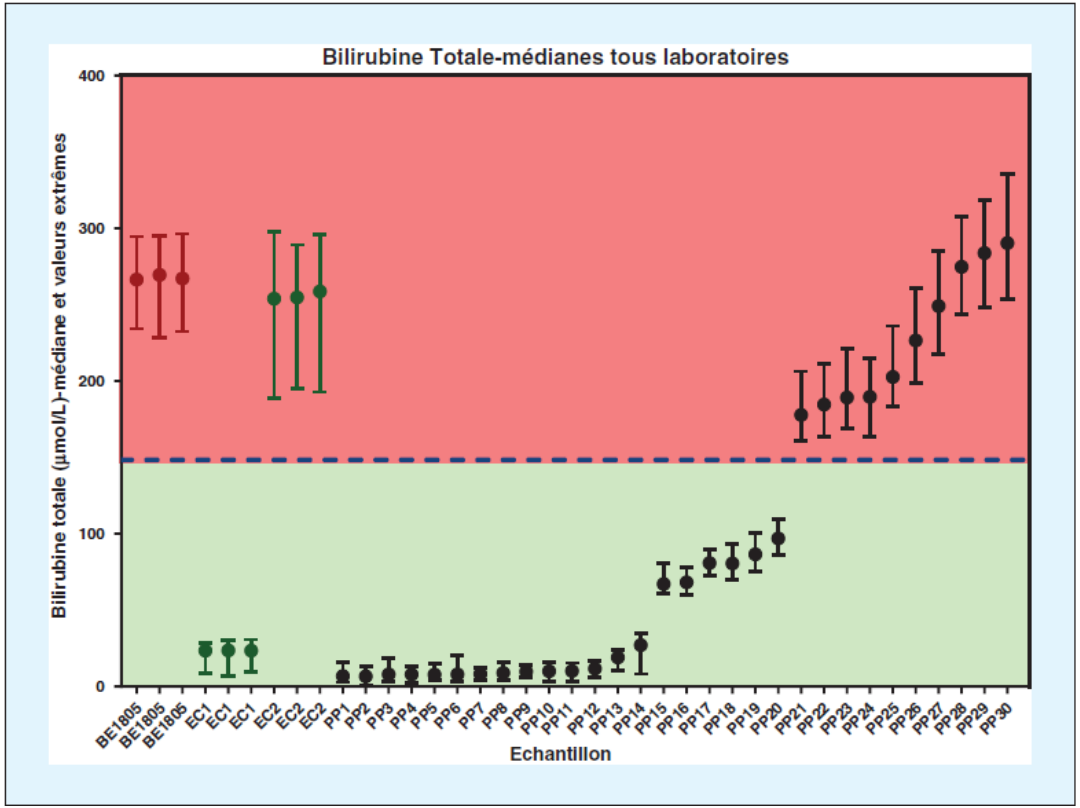


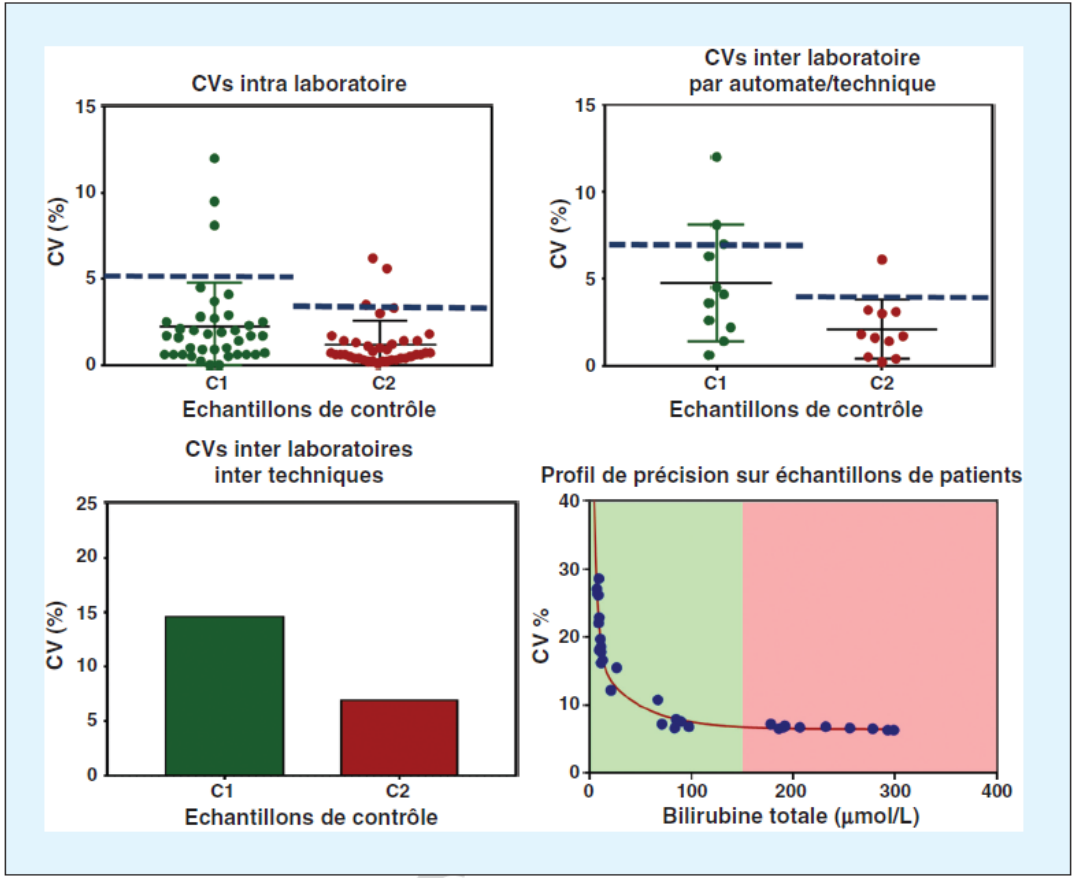
Figure 3. Répartition des LBM participant à l'étude d'harmonisation, par type de LBM (**A**) et par automates utilisés (**B**).

Fournisseur	Automate	Méthode	LBM	Effectif
Abbott	Alinity	Autres diazoïques	1-6-30-42	4
	Architect	Dichloraniline + solubilisant	8	5
Beckman	AU	Diphényldiazonium (DPD) + solubilisant	31	3
	DXc	Autres diazoïques	21	1
Ortho	Vitros TBIL	Réflectométrie	37, 51	2
	Vitros BuBc	Réflectométrie	37	1
Roche	Cobas	Diphényldiazonium (DPD) + solubilisant	2-10-11-12-13-14-15-16-18-24-28-32-34-39-43	15
Siemens	Advia	Vanadate oxydation	5-36	2
	Atellica	Autres diazoïques	49-50	2
	Vista	Acide sulfanilique + caféine-benzoate "rose"	17-19-33	3
ThermoFisher	Indiko	Autres diazoïques	26	1

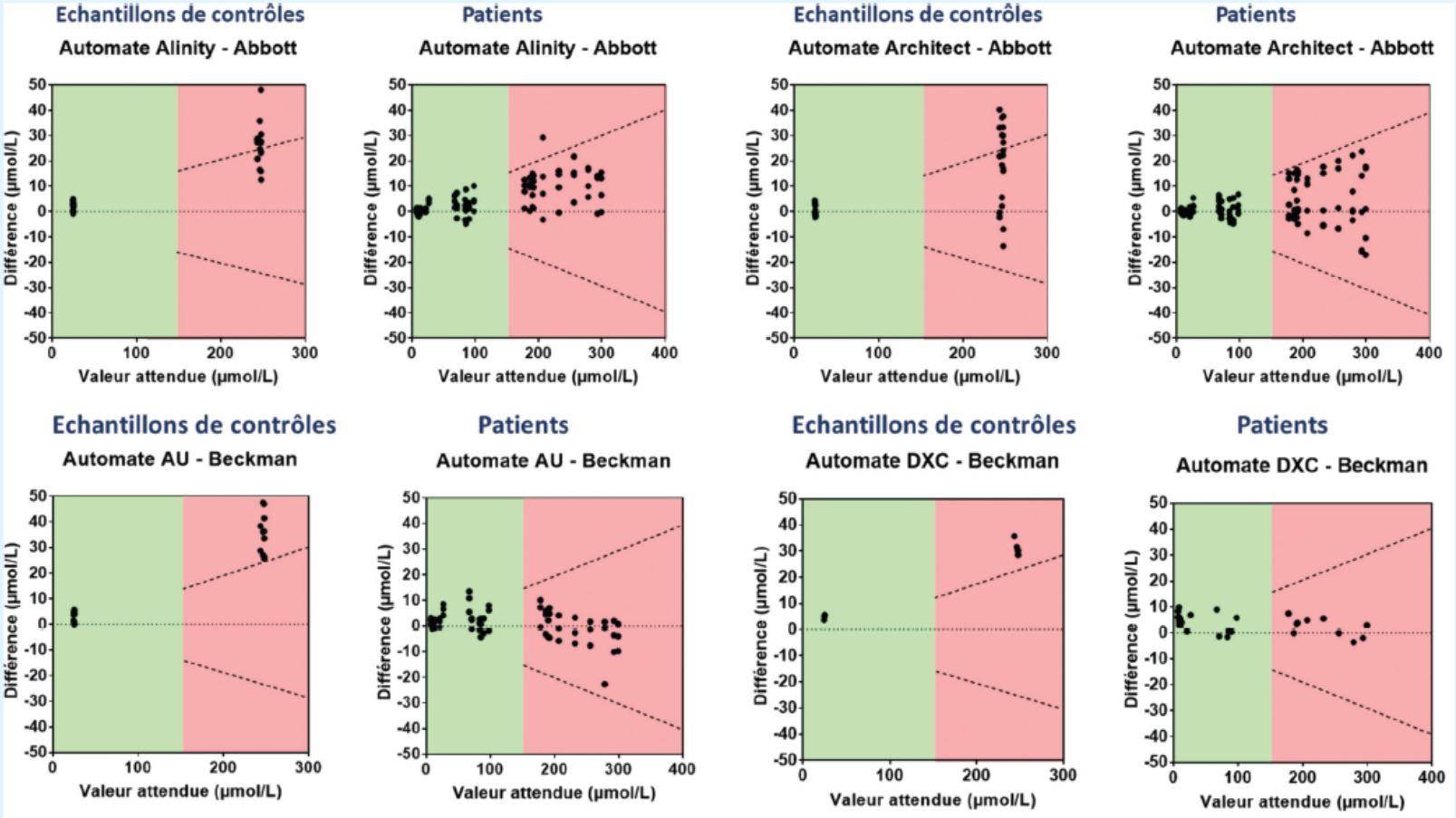
Results at a glance



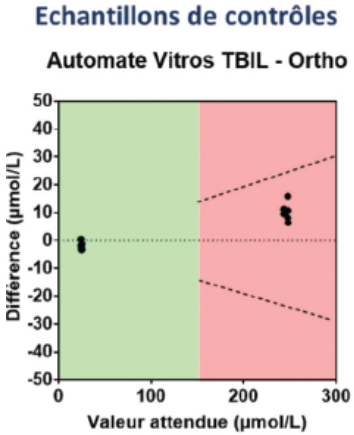
Imprecision

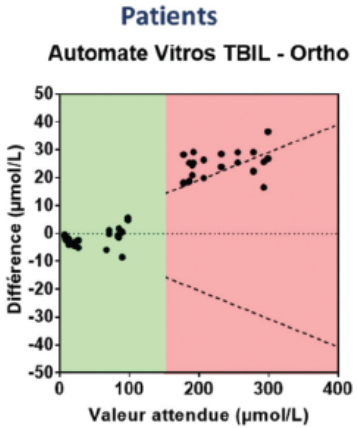


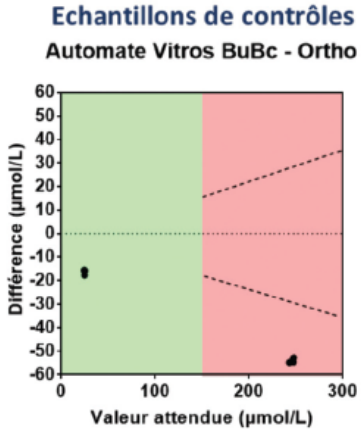
Bias (1)

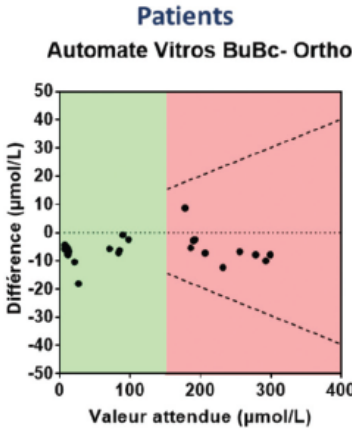

Echantillons de contrôles
Automate Alinity - Abbott
Patients
Automate Alinity - Abbott
Echantillons de contrôles
Automate Architect - Abbott
Patients
Automate Architect - Abbott
Echantillons de contrôles
Automate AU - Beckman
Patients
Automate AU - Beckman
Echantillons de contrôles
Automate DXC - Beckman
Patients
Automate DXC - Beckman
Différence (µmol/L)
Valeur attendue (µmol/L)

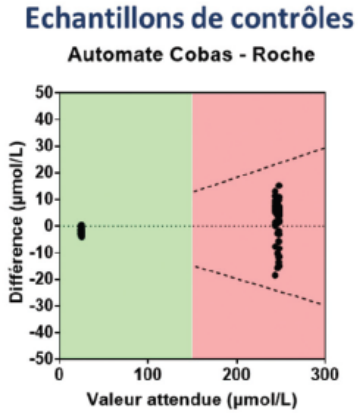
Bias (2)

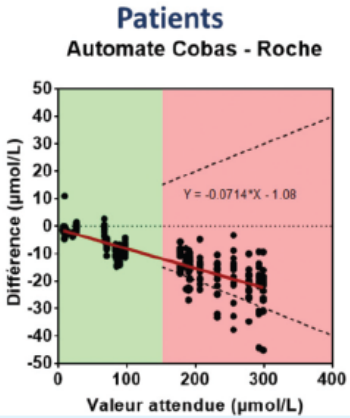

Echantillons de contrôles
Automate Vitros TBIL - Ortho
Différence (µmol/L)
Valeur attendue (µmol/L)

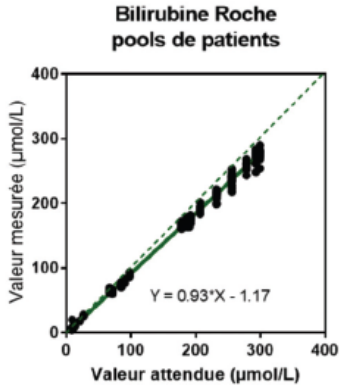

Patients
Automate Vitros TBIL - Ortho
Différence (µmol/L)
Valeur attendue (µmol/L)


Echantillons de contrôles
Automate Vitros BuBc - Ortho
Différence (µmol/L)
Valeur attendue (µmol/L)


Patients
Automate Vitros BuBc- Ortho
Différence (µmol/L)
Valeur attendue (µmol/L)


Echantillons de contrôles
Automate Cobas - Roche
Différence (µmol/L)
Valeur attendue (µmol/L)


Patients
Automate Cobas - Roche
Y = -0.0714*X - 1.08
Différence (µmol/L)
Valeur attendue (µmol/L)


Bilirubine Roche
pools de patients
Y = 0.93*X - 1.17
Valeur mesurée (µmol/L)
Valeur attendue (µmol/L)

French National multicenter study CNRHP – SFBC – CNBH
Harmonization between French labs (3) – Accuracy

- The methods used in France present significant inter-technical differences, linked to a drift in standardization over time (SRM916a).

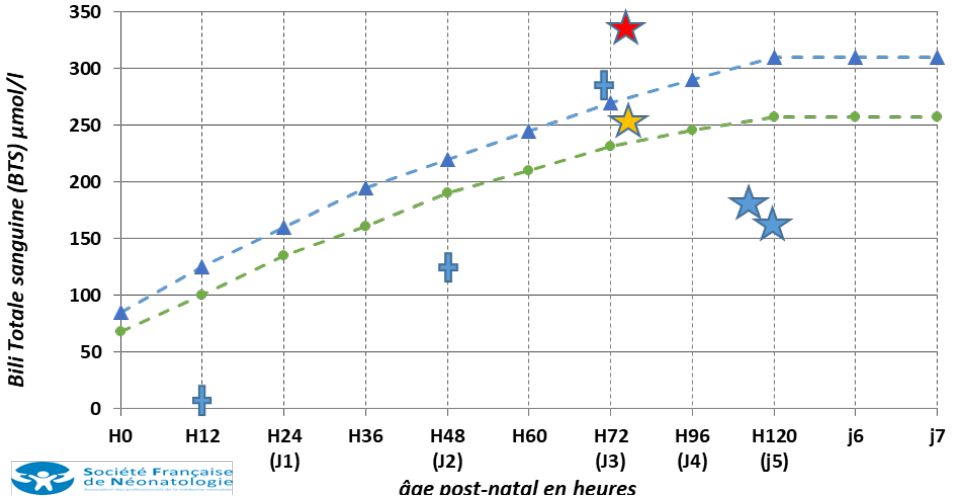
- Above 150 µmol/L, there are deviations of more than 20% from expected values that may influence the clinical decision:
  - Either an underestimation with the Roche method (the most widespread in France): possible clinical impact with possible delay in management
  - ✓ Either an overestimation with less clinical impact: phototherapy initiated incorrectly or more quickly

However the existence of bias with respect to the reference value was observed

IVD company	Analyzer	Relative bias for patients >150 µM
Abbott	Alinity	+ 10
	Architect	+ 20 (dispersion)
Beckman	AU	0
	DXc	+ 5
Ortho	Vitros TBIL	+ 30 (clinical incidence)
	Vitros BuBc	- 10
Roche	Cobas	- 25 (clinical incidence but possible correction *in silico)*
Siemens	Advia	+ 15
	Atellica	+ 20
	Vista	+ 5
ThermoFisher	Indiko	+ 10

French National multicenter study CNRHP – SFBC – CNBH
Harmonization between French labs (4) – Clinical incidence

Born at 37 GW / mother with blood group A RhD pos and RAI negative.
Good adaptation. Exclusive breastfeeding with difficulty at the beginning.
Weight loss at H72 12%
No early jaundice but severe jaundice at H72 with TcB at 270 µmol/l + motivating TSB locally at 264 µmol/l ★ and at CNRHP at 302 µmol/l ★



The clinical impact of an underestimation of serum bilirubin was reported in a case at the Armand Trousseau Hospital with delay in management. Requests for advice from the CNRHP confirmed that this type of situation occurs regularly in other centers.

Method performance (in 2019)

Tableau 4. Synthèse des résultats de l'évaluation de l'inexactitude.

Fournisseur	Automate	Contrôle C1	Contrôle C2	Patients < 150 µmol/L	Patients > 150 µmol/L
Abbott	Alinity	A	R+ 25	A	A+ 10
	Architect	A	R+ 20	A	A+ 20 (dispersion)
Beckman	AU	A	R+ 35	A	A
	DXc	A	R+ 30	A	A+ 5
Ortho	Vitros TBIL	A	A+ 10	A	R+ 30 (pas d'incidence clinique)
	Vitros BuBc	A	R-35	A	A- 10
Roche	Cobas	A	A	A	R- 25 (correction possible)
Siemens	Advia	A	A+ 15	A	A+ 15
	Atellica	A	A+ 20	A	A+ 20
	Vista	A	A+ 5	A	A+ 5
ThermoFisher	Indiko	A	A+ 15	A	A+ 10

A : accepté, R : rejeté, vs objectifs analytiques - + ou – XX : biais moyen en µmol/L.

Calibration strategy

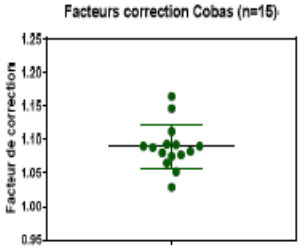
- Possibility to cover concentrations between 50 and 400 µM with a 2-point calibration (one point slightly above 50 µM and another between 250 and 400 µM) but low values not covered
- With 2 points, need to choose between low and intermediate values
- To cover low values, 3-point calibration is necessary with a low point around 25 µM, a second slightly above 50 µM and a third between 250 and 400 µM.
- Selection of the standard around 25 µM difficult without guarantee of improvement of the accuracy at low values: improvement of methods necessary
- Standardization of calibration insufficient to improve the accuracy of routine methods for low values but improvement of inter-technical comparability is expected.
- For high values, "standardization" of the calibration should allow an improvement of both the accuracy and the comparability of the results.
- Possibility to use HANL, HANH and BNLAEG samples to evaluate the accuracy of methods for which they are commutable.

Method performance (in 2019) after in silico recalibration

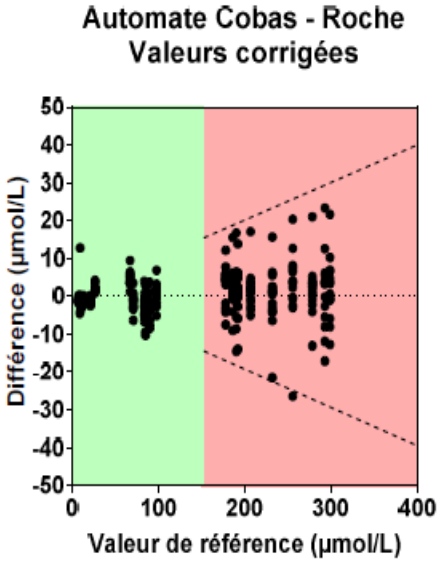
Exemple de correction (µmol/L)
- ✓Valeur PP30 LBM = 289,6
- ✓Moyenne BE1805 LBM = 257,8
- ✓Valeur assignée BE1805 = 270,6
- ✓**Valeur corrigée PP30 LBM = 289,6*270,6/257,8** = 303,1

Exactitude – Biais vs. Valeurs de références
Après correction sur Roche – Cobas

Facteur de correction calculé pour chaque LBM à partir de la moyenne des valeurs obtenues pour l'étalon BE1805
F = 1,09 ± 0,03 (1,03 – 1,16)



Limite d'acceptabilité clinique : 10 % pour des valeurs > 150 µmol/L
Exactitude sur échantillons de patients aux seuils de décision en néonatalogie : accepté après correction *in silico* (en 1 point vs valeur BE1805)



French National multicenter study CNRHP – SFBC – CNBH
Harmonization between French labs (4) – Conclusion

This analytical study of multicenter harmonization allowed to:

- **better specify the nature and intensity of inter-technical variations i**n the measurement of total bilirubin concentrations in neonatology, according to the techniques used
- **propose simple measures** to correct the results when consequences on the clinical decision may occur with a direct application of the interpretation criteria recommended societies (SFN).