Comprehensive, Near Patient Hyperbilirubinemia Testing in Newborns Using Low Blood Volume

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INTRODUCTION

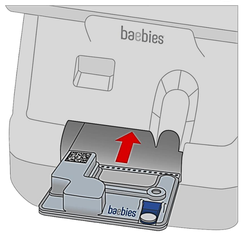
- Neonatal jaundice is common and can lead to bilirubin induced neurotoxicity (BIND), kernicterus and/or other severe complications if not treated promptly.
- Comprehensive risk assessment for bilirubin toxicity requires testing for multiple analytes including total serum bilirubin (TSB), albumin, glucose-6-phosphate dehydrogenase (G6PD) enzyme activity and bilirubin binding capacity (BBC).
- Currently each of these tests is performed separately, some require send-out with long turnaround times and BBC is not commercially available.
- Prolonged time to result and lack of access to BBC testing contribute to hospital readmissions and suboptimal care of neonates with severe hyperbilirubinemia.

OBJECTIVE

To demonstrate laboratory-equivalent, clinically-actionable measurements of TSB, albumin and G6PD from a single drop of whole blood using a benchtop rapid analyzer with a run time of less than 15 minutes.

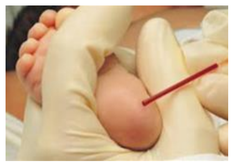
METHODS

Insert Cartridge



Microfluidic cartridge with multi-test capability

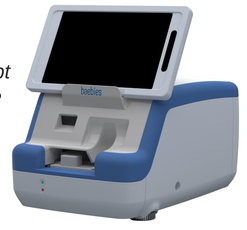
Add Sample



50 µl whole blood input

Run Test

Concept device



Approximate run time of 15 minutes for all 3 tests, including on-board plasma separation

Traditional Hyperbilirubinemia Test Methods

TSB	Albumin	G6PD	Cumulative
minutes	minutes	hours	hours
500 µl	500 µl	500 µl	> 1.5 mLs

Near Patient Hyperbilirubinemia Testing

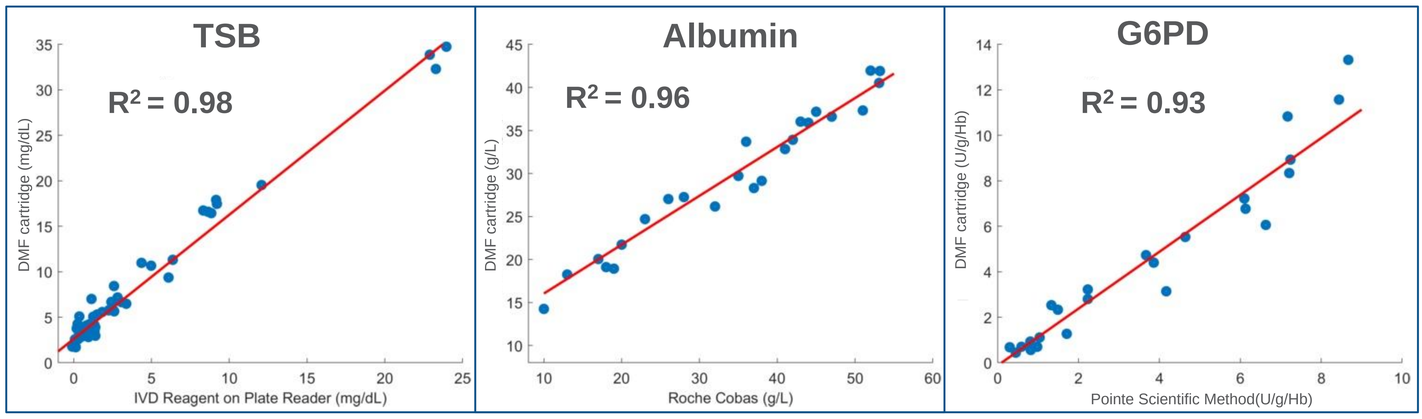
All Analytes
< 15 minutes
< 50 µl

RESULTS

SUMMARY OF RESULTS

Analyte	Method	Assay Format	Range Tested	Method Comparison R^2
TSB	Diazo	Endpoint colorimetric	up to 35.0 mg/dL	0.98
Albumin	BCG	Endpoint colorimetric	up to 41.0 g/L	0.96
G6PD	NADPH	Kinetic fluorimetric	up to 8.7 U/g/Hb	0.93

CLINICAL METHOD COMPARISON



Method comparisons of standard clinical test methods (X-axis) to the near patient digital microfluidic cartridge assays (Y-axis) for TSB, albumin and G6PD. The red line represents the linear fit of all data points. For all assays, the DMF results compare well to the gold standard laboratory values.

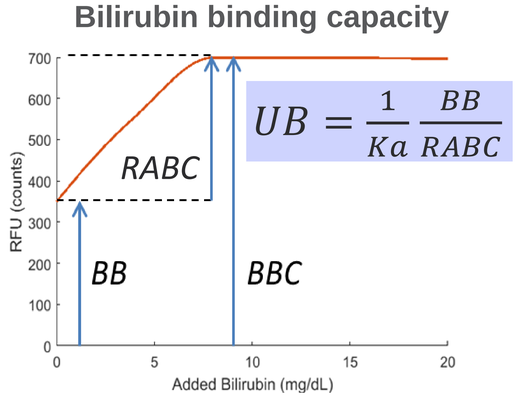
CONCLUSIONS

- **These results demonstrate that laboratory-equivalent values for TSB, albumin and G6PD enzyme activity can be obtained using a small footprint rapid analyzer with a total run time of under 15 min.**
- **The system uses digital microfluidic (DMF) technology to automate kinetic fluorimetric enzyme assays (G6PD) and dye binding colorimetric assays (TSB, albumin) simultaneously from a single drop (<50 µl) of whole blood.**
- **All reagents, including plasma separation chemicals, are stored on-board the disposable cartridge; the system is suited to a variety of clinical settings including clinics and hospitals.**

FUTURE DIRECTIONS

Additional near patient tests are currently in active development both to complement the hyperbilirubinemia test panel (BBC, unbound bilirubin, etc.) and to address other time critical neonatal conditions (metabolic disorders, hypercoagulation, etc.).

Bilirubin binding capacity



An assay for BBC, an indirect measure of unbound bilirubin (UB) or the capacity of albumin to bind bilirubin, was adapted to the DMF platform (left). Briefly, the fluorescence signal of albumin-bound bilirubin (red line) in a single sample droplet was measured kinetically following serial additions of 0 mg/dL to 20 mg/dL of dried bilirubin (X-axis). BB: endogenous bound bilirubin in the sample; RABC: reserve albumin binding capacity; K_a: association constant of albumin.

ACKNOWLEDGEMENTS

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