scientific reports

Check for updates

OPEN

Performance evaluation of the BactInsight simplified blood culture system developed for resource-limited settings using a simulated test design

Barbara Barbé[1,5]✉, Jens Cornelis[1,2,3,5], Mohammadamin Ghomashi[2,3], Ellen Corsmit[1], Els Genbrugge[1], Federico Marchesin[2,3], Yanlu Li[2,3], Roel Baets[2,3], Jan Jacobs[1,4] & Liselotte Hardy[1]

BactInsight is a simplified blood culture system designed for resource-limited settings. It consists of in-house produced blood culture bottles and a turbidimeter to detect microbial growth on top of visual inspection of blood culture bottles. A first proof-of-concept using simulated commercial blood cultures showed that the turbidimeter was able to detect growth in nine out of ten tested microbial species (i.e. by growth detection through turbidity and a colour indicator), however detection of turbidity was only successful in four of these species. In this study, we extended the in vitro testing to 20 microbial species spiked in fresh human blood and we used a second-generation turbidimeter prototype and in-house produced blood culture bottles. The BactInsight system's performance was acceptable when compared with the automated BACT/ALERT system (bioMérieux). Moreover, the addition of the turbidimeter to visual inspection decreased the time-to-positivity for some microorganism groups. The system is cheap (~ 50 USD), robust and able to withstand high temperature and humidity. In conclusion, the BactInsight system has the potential to improve access to blood cultures in resource-limited settings.

Blood cultures are the diagnostic of choice to detect bloodstream infections, despite their low sensitivity and long turn-around time[1–3]. The World Health Organisation (WHO) confirmed this by declaring blood cultures a priority specimen for antimicrobial resistance surveillance[4]. In high-income settings, automated blood culture systems which incubate, agitate, and continuously monitor growth in blood culture bottles (BCBs) are part of the standard of care[5,6]. When growth is detected, based on CO_2-production in the BCB, a signal is given by the automate.

Automated systems and their respective BCBs are expensive and require regular maintenance and uninterrupted power supply. In resource-limited settings (RLS), these requirements are often not met. In addition, clinical bacteriology laboratories are scarce in these settings[7,8]. If a clinical bacteriology laboratory is available, manual blood culture systems are mostly used: BCBs are incubated in a conventional incubator at 35–37 °C with daily visual inspection for growth. Although more affordable, these manual systems have a lower yield and a longer time-to-positivity (TTP) than automated systems, and visual detection is subjective and depends on the expertise of the end-user[1].

We developed a simplified blood culture system "BactInsight" (following the target product profile developed for RLS[9], consisting of in-house produced BCBs and a reader ("turbidimeter") intended to complement, objectivize, and accelerate visual inspection. In addition, the system is cheap (~ 50 USD), robust and able to withstand high temperature and humidity. We previously reported the pilot phase results of the first-generation turbidimeter[10]. Based on these first results, including both its successes and shortcomings, we improved the design and functionalities into a second-generation prototype: the new prototype is built using an improved

[1]Department of Clinical Sciences, Institute of Tropical Medicine, Antwerp, Belgium. [2]Center for Nano- and Biophotonics (NB-Photonics), Ghent University, Ghent, Belgium. [3]Photonics Research Group, Department of Information Technology, Ghent University-IMEC, Ghent, Belgium. [4]Department of Microbiology, Immunology and Transplantation, KU Leuven, Leuven, Belgium. [5]Barbara Barbé and Jens Cornelis contributed equally to this work. ✉email: bbarbe@itg.be

version of the hard- and software, has a modular design and enhanced turbidity detection. In this study, we evaluated the performance of the BactInsight system using the BACT/ALERT 3D system (bioMérieux, Marcy-l'Etoile, France) as a reference system. As an exploratory objective, we assessed the added value of the turbidimeter for a manual blood culture system.

Methods

In-house produced blood culture bottles

Polycarbonate BCBs with a total volume of 70 ml (Zhuhai Ideal Biotech Co., Ltd, Guangdong, China) were filled with tryptic soy broth (TSB; Merck, Burlington, Massachusetts, USA) (30 ml) supplemented with sodium-polyanethole sulfonate (SPS; Merck) (0.3 mg/ml) and capped with a rubber septum and an aluminium crimp top seal (Fisher Scientific, Waltham, MA, USA). Next, the prepared BCBs were autoclaved for 15 min at 121 °C.

Second-generation turbidimeter prototype: concept and design

The turbidimeter is a small growth-detection device (58 mm x 81 mm x 141 mm) built with low-cost, off-the-shelf components, fitted into a custom-made 3D-printed casing. The following adaptations were made to the previously described design[10] (Fig. 1): (i) The Arduino development board was replaced by a nRF52840-based PAN1780 microcontroller (Panasonic, Osaka, Japan), allowing for more advanced and reliable software to control the turbidimeter. (ii) The 598 nm (amber) LED was replaced by a brighter 4000 K LED (Luminus, Sunnyvale, USA). (iii) The silicon photodiode was replaced by two TCS3772 RGBC-sensors (ams-OSRAM, Premstaetten, Austria), one to measure transmission and one to measure scattering of light. (iv) A modular sensing system was implemented: transmitted and scattered light were measured by two separate, but identically designed detection daughterboards, while a third emitter daughterboard housed the LED. The transmission detector board was mounted opposite (180°), and the scattering detector board perpendicular (90° angle) to the emitter board. (v) As the in-house produced BactInsight BCBs did not have a colour indicator at the bottom (as opposed to the commercial BCBs used to evaluate the first-generation turbidimeter), the red-green-blue (RGB) colour sensor at the base of the turbidimeter was removed. The total bill of materials (BOM) cost of the second-generation turbidimeter was around 50 USD (equal to the first-generation prototype), when ordering the components in large volumes (i.e. to produce 1000 turbidimeter modules). The turbidimeter was developed to be maintenance-free, and in case of technical problems, the components, such as the three daughterboards, can easily be replaced by the end-user.

Spiking of the blood culture bottles

Human heparinized blood collected from eight healthy volunteers (ethical approval 27/2022, institutional review board of the Institute of Tropical Medicine) was spiked with reference and clinical strains belonging to 20 clinically relevant microbial species (Table 1). The human blood was tested for sterility by including negative control BCBs (i.e. inoculated with non-spiked blood) and was controlled for by extensive blood testing (including white blood cell count, haematocrit, and haemoglobin tests). In-house produced BCBs were inoculated as described previously[10–12] (Fig. 2). The final concentration in the BCB was ~ 15 colony forming units (CFU)/ml blood. This was verified by inoculating the final dilution (100 µl) onto three blood agar plates for colony count (acceptable range 10–100 CFU/100 µl). Runs with a colony count outside of this acceptable range were excluded from analysis, i.e. only runs with a final concentration of 4–40 CFU/ml blood were included in the data analysis. All healthy volunteers provided informed consent to use their samples for this study.

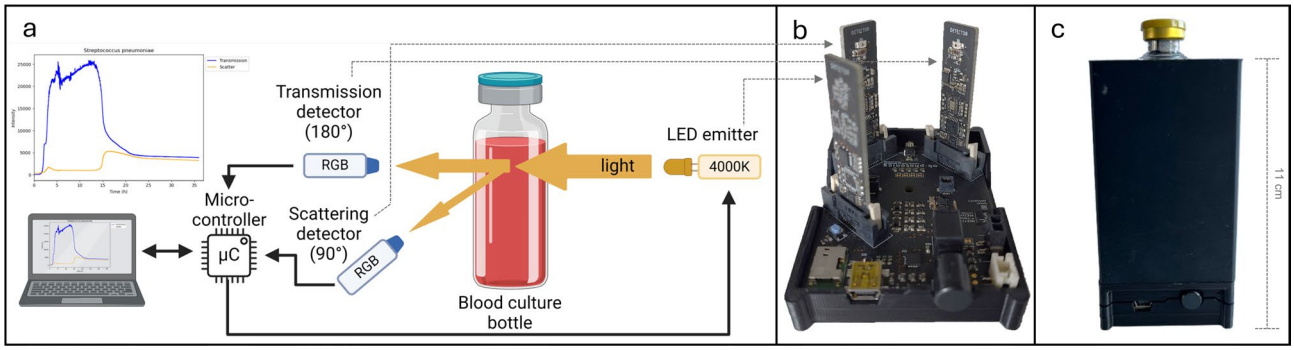

Fig. 1. Turbidimeter concept and design. (Panel **A**) A 4000 K light emitting diode (LED) on the emitter daughterboard illuminates the blood culture broth every 30 s. The detector daughterboards capture the light after it has passed through the broth: transmitted light (180°) and scattered light (90° angle) are detected. Raw sensor data are transferred in real time to a computer through a USB connection in a CSV format. A logging program on the computer stores this information in a text-file and a custom-made algorithm is used for growth detection. Created in BioRender. Cornelis, J. (2025) https://BioRender.com/1lz0so3. (Panel **B**) Picture of the opened turbidimeter module, showing the three daughterboards (1 emitter and 2 detector boards) mounted in a modular format onto the motherboard. (Panel **C**) Picture of the turbidimeter module mounted into the 3D-printed casing. The blood culture bottle is inserted into the holder which is closed off by a lid.

Organism group	Species	*N* of strains tested		Suspension (McF)	Dilution factor	Incubation time (hours)
		Reference	Clinical			
Enterobacterales	*Escherichia coli*	1	6	0.5	400,000	20–48
	Salmonella Typhimurium	1	6	0.5	400,000	20–48
	Klebsiella pneumoniae	1	5	0.5	400,000	36–48
	Salmonella Typhi	1	5	0.5	400,000	20–48
	Enterobacter cloacae	1	6	0.5	200,000	36–96
Non-fermenters	*Pseudomonas aeruginosa*	1	6	0.5	400,000	36–48
	Acinetobacter baumannii	1	6	0.5	200,000	48
	Burkholderia cepacia	1	0	0.75	400,000	96
Staphylococci	*Staphylococcus aureus*	1	15	0.5	400,000	36–48
	Staphylococcus epidermidis	1	5	0.5	150,000	48–96
Streptococci	*Streptococcus pneumoniae*	1	6	1.0	150,000	36–48
	Streptococcus pyogenes	1	6	0.5	400,000	36–48
	Streptococcus anginosus	1	5	0.5	150,000	36–48
	Streptococcus suis	1	6	0.5	600,000	36–48
Enterococci	*Enterococcus faecalis*	1	6	0.5	400,000	36–48
Fastidious organisms	*Haemophilus influenzae*	1	0	0.5	400,000	96
	Neisseria subflava	1	0	0.5	400,000	96
Yeast	*Candida albicans*	1	0	0.5	10,000	96
	Candida tropicalis	1	0	0.75	10,000	96
	Cryptococcus neoformans	1	0	0.75	8,000	96
	TOTAL	**20**	**89**			
		109				

Table 1. List of the 20 clinically relevant microbial species selected for testing. McF = MacFarland, *N* = number.

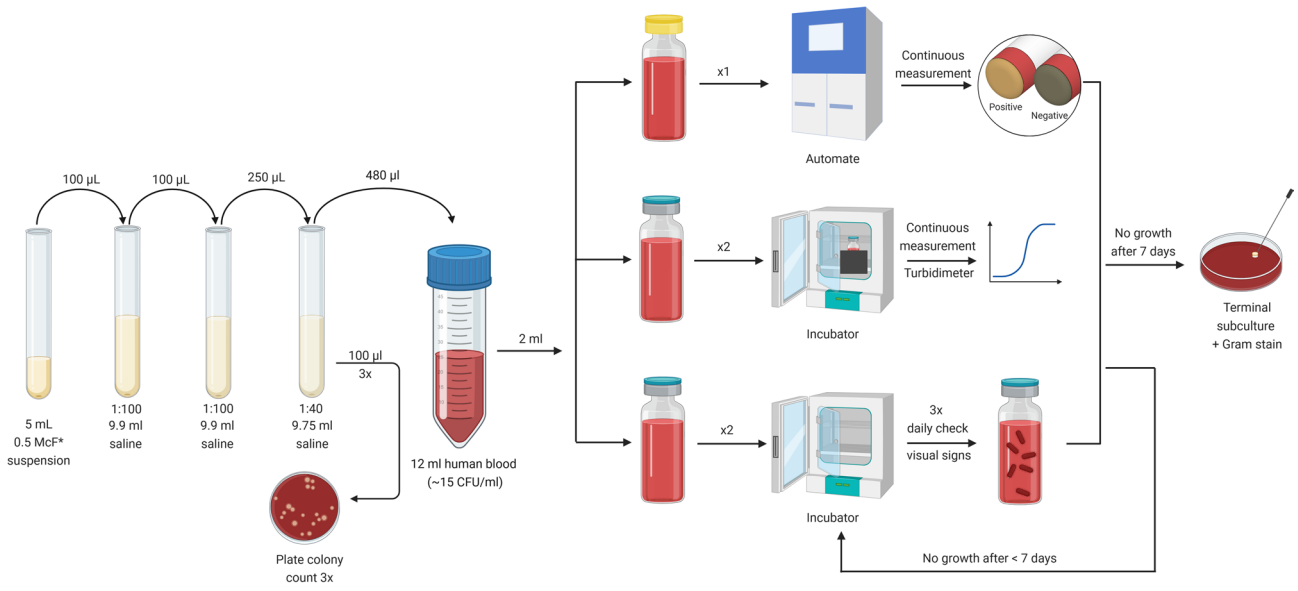

Fig. 2. Overview of the simulated test design with spiked blood culture bottles. The spiking of the blood culture bottles (BCBs) was done similarly as previously described[10,12]: a suspension of 20 microbial species was serially diluted to a final concentration of 15 CFU/ml fresh human heparinized blood, next 2 ml of the spiked blood was inoculated per BCB. For each strain to be tested, four BactInsight BCBs and one BACT/ALERT PF Plus bottle (bioMérieux) were inoculated. The reference bottle was placed in the BACT/ALERT 3D 120 (bioMérieux) automate (continuous agitation and growth monitoring every 10 min). Two BactInsight BCBs were placed in turbidimeter modules in a conventional incubator (no agitation, measurements every 30 s), the two remaining BactInsight BCBs were placed in a conventional incubator and inspected for growth three times per day. After incubation, the BCBs were subcultured on blood agar plates to check for purity. Created in BioRender. Barbé, B. (2025) https://BioRender.com/hn55eit.

Simulated test design with spiked blood culture bottles

For each strain to be tested, four BactInsight BCBs and one BACT/ALERT PF Plus bottle (bioMérieux) were inoculated (Fig. 2). Two BactInsight BCBs were placed in the turbidimeter modules inside an incubator, with measurements done every 30 s. The two remaining BactInsight BCBs were incubated and visually inspected for growth three times per day. Incubation was done in a conventional incubator at 35–37 °C, without agitation, for the defined incubation time (Table 1). The BACT/ALERT PF Plus bottle was incubated in the BACT/ALERT 3D 120 automate (bioMérieux) at 35 °C, with continuous agitation and growth monitoring every 10 min. After incubation, the BCBs were subcultured on blood agar plates to confirm growth and check for purity.

Definitions

A run was defined as an experiment whereby one microbial strain was inoculated into five different BCBs (four BactInsight BCBs and one reference BCB) and tested with three different systems. A set of BCBs is the combination of two BactInsight BCBs, the first measured by turbidimeter and the second by visual inspection.

Yield was defined as the percentage of spiked BCBs with growth detected by the system on the total number of bottles with confirmed growth (by subculture). For the reference system, growth detection was defined as a positive signal given by the automate. For the BactInsight system, growth detection by either visual inspection and/or the turbidimeter was interpreted as a positive signal for growth.

TTP was defined as the delay between incubation and detection of growth. For the reference system, the TTP was automatically provided by the automate. For the BactInsight system, the TTP was the shortest TTP provided either through visual inspection or the turbidimeter for each set of BCBs.

Specificity of the turbidimeter was defined as the percentage of negative control BCBs with no growth detected by the turbidimeter on the total number of negative control BCBs.

Data analysis

Data entry was done in the REDCap web platform (Vanderbilt University) installed on a tablet. Turbidimeter data were exported in csv files and analyzed using a custom-made growth algorithm in Python which assessed the relative change in the intensity of transmitted or scattered light after having passed through the BCB broth. The growth detection algorithm was defined by a decrease in transmitted light by at least 15% or an increase in scattered light by at least 50%. The first 8 h of the measurement were not taken into account to allow the sedimentation of the blood in the BCB. In addition, peaks and dips were filtered out by convolving the obtained growth curves with a median kernel of length seven. This length was chosen so that the biggest artefacts in the measurement curves were filtered out.

Data analysis was performed in R version 4.1.2 (R Foundation for Statistical Computing, Vienna, Austria). To compare the BactInsight system performance with the reference system, overall yield was calculated and the median TTP (with quartiles 1–3 (Q1-Q3) and min/max range) was calculated per system, overall and per species. Further, a descriptive analysis of the turbidimeter's performance was done based on the yield and TTP of the turbidimeter, as described above, and compared to the results of the visual inspection using a Wilcoxon signed rank test (paired data).

All methods were performed in accordance with the relevant guidelines and regulations.

Results

A total of 212 runs were conducted between November 2022 and June 2023 across both the BactInsight and reference systems, of which 43 runs were excluded for various reasons (mainly technical or procedural issues, Supplementary Figure S1). In the end, a total of 169 runs were withheld for analysis, consisting of 502 BCBs (i.e. 333 sets of BactInsight BCBs and 169 reference BCBs) inoculated with 109 different strains belonging to 20 microbial species (Supplementary Table S2). Turbidimeter detection failed for yeast and fastidious organisms ($n = 28$), therefore the turbidimeter results were not taken into account for these species and only the visual inspection results were withheld for analysis.

Performance of the BactInsight system, compared with the reference system

Growth was detected in all 502 BCBs resulting in a yield of 100% for both the BactInsight (333/333) and the BACT/ALERT system (bioMérieux) (169/169).

The median BactInsight TTP (14.93 h; Q1-Q3 11.51–20.22 h) was 1.49 h longer than the median automate TTP (13.44 h; Q1-Q3 12.72–16.56 h). The BactInsight TTP ranged from 9.39 to 69.65 h compared to a range of 10.56 to 57.12 h for the reference automate (Supplementary Figure S3). Day 1 growth (TTP ≤ 24 h) was observed in 89.8% (299/333) of BactInsight BCB sets versus 95.9% (162/169) of reference BCB sets, day 2 growth (TTP ≤ 48 h) in 95.2% (317/333) versus 98.2% (166/169) respectively, and day 3 growth (TTP ≤ 72 h) was 100% (502/502) for both systems.

The median BactInsight TTPs were shorter (<-1 h) than the reference system in 6/20 (30.0%) species tested (i.e. most Enterobacterales, enterococci and some streptococci). Both systems had similar TTPs (i.e. difference in median TTP of maximum one hour) for 3/20 (15.0%) species tested (i.e. most streptococci and *Pseudomonas aeruginosa*). For 11/20 (55.0%) species tested, the median BactInsight TTPs were longer (> 1 h) than those of the reference system. This was the case for most non-fermenters and staphylococci, but the largest TTP differences and variations were observed for fastidious organisms and yeasts (Table 2; Fig. 3).

Performance of the turbidimeter only and comparison with visual inspection

The overall turbidimeter yield was 97.4% (297/305): 98.5% (128/130) for reference strains, and 96.6% (169/175) for clinical strains (Supplementary Table S4). Yield below 100% (reference and clinical strains combined) was observed for *Acinetobacter baumannii* (70.6%, 12/17), *Streptococcus anginosus* (90.0%, 18/20) and *Staphylococcus*

Pathogen group	Species	TTP (hours) BactInsight system				TTP (hours) BACT/ALERT system (bioMérieux)				Difference in median TTP (hours)
		N	Median	Min-Max	Q1-Q3	*N*	Median	Min-Max	Q1-Q3	
Enterobacterales	*Escherichia coli*	18	10.52	9.39–13.52	10.36–10.77	10	12.00	11.76–12.24	11.82–12.24	-1.48
	Salmonella Typhimurium	22	10.71	10.19–11.46	10.58–10.90	11	12.72	12.24–14.64	12.48–12.84	-2.02
	Klebsiella pneumoniae	20	16.34	10.30–20.80	13.15–18.27	10	12.00	10.56–12.48	11.82-12.00	4.34
	Salmonella Typhi	18	12.71	11.59–14.22	12.41–12.81	10	14.88	14.16–15.84	14.46–15.36	-2.17
	Enterobacter cloacae	24	11.45	10.29–16.67	11.01–11.89	12	12.96	12.48–14.40	12.72–12.96	-1.51
Non-fermenters	*Pseudomonas aeruginosa*	20	18.03	15.71–20.57	17.17–18.32	10	17.28	16.08–19.68	16.68–18.24	0.75
	Acinetobacter baumannii	17	20.57	14.91–20.85	16.43–20.75	9	12.24	11.28–13.68	12.00-13.20	8.33
	Burkholderia cepacia	6	24.07	18.82–37.58	20.13–33.24	3	20.88	20.16–20.88	20.52–20.88	3.19
Staphylococci	*Staphylococcus aureus*	36	18.42	15.00-25.83	16.55–21.36	18	14.52	12.72–19.44	13.92–15.12	3.90
	Staphylococcus epidermidis	20	21.64	19.02–31.28	21.58–21.97	10	16.68	14.40-17.76	16.20-16.98	4.96
Streptococci	*Streptococcus pneumoniae*	20	14.27	11.61–17.54	12.42–14.97	10	13.44	12.48–16.56	12.78–13.44	0.82
	Streptococcus pyogenes	20	12.23	10.15–17.03	11.61–13.68	10	13.20	12.00-14.16	12.54–13.80	-0.98
	Streptococcus anginosus	20	19.57	12.51–45.30	15.29–22.12	10	18.48	17.28–32.64	18.06–23.04	1.09
	Streptococcus suis	22	11.29	10.12–12.42	10.91–11.65	11	12.96	12.24–13.20	12.48–13.20	-1.67
Enterococci	*Enterococcus faecalis*	22	11.58	9.91–20.35	10.61–12.41	11	12.96	11.52–13.44	12.72–13.32	-1.38
Fastidious organisms	*Haemophilus influenzae**	4	56.22	42.83–69.60	42.83–69.60	2	20.88	20.16–21.60	20.52–21.24	35.34
	*Neisseria subflava**	6	65.00	18.85–69.58	30.39–68.44	3	16.80	16.08–17.04	16.44–16.92	48.20
Yeast	*Candida albicans**	6	42.85	20.93–65.37	31.71–59.74	3	26.64	22.80–27.60	24.72–27.12	16.21
	*Candida tropicalis**	6	65.00	28.02–69.65	37.26–68.49	3	20.16	19.92–20.40	20.04–20.28	44.84
	*Cryptococcus neoformans**	6	65.00	28.00-69.60	37.25–68.45	3	55.20	48.72–57.12	51.96–56.16	9.80

Table 2. Overview of the time-to-positivity observed per species and per system. Note that the TTP result of one set of BCBs was manually corrected (*Streptococcus pneumoniae* M0256). Min-Max = minimum-maximum (range), *N* = number, Q1-Q3 = quartile 1 - quartile 3, TTP = time-to-positivity. * TTP of the BactInsight system only takes into account visual inspection.

aureus (97.2%, 35/36). A total of 41 negative control BCBs were tested with the turbidimeter, of which 14.6% (6/41) were detected as positive (i.e. 85.4% specificity).

The turbidimeter TTP ranged from 9.39 to 55.10 h (Q1-Q3 11.32–21.26 h), with a median TTP of 13.67 h, which was lower than that of the visual inspection (median TTP of 21.03 h, range 15.00-69.62, Q1-Q3 18.47–21.93) (Supplementary Figure S5). The TTP results obtained by the turbidimeter and visual inspection were compared by means of a Wilcoxon signed-rank test (Supplementary Table S6). The overall TTP of the turbidimeter was shorter than the overall visual inspection TTP (p-value < 0.0001). The turbidimeter TTPs were shorter for Enterobacterales, streptococci (except *Streptococcus anginosus*) and enterococci (p-values < 0.05), while the visual inspection TTPs were shorter for *Acinetobacter baumannii* and *Staphylococcus aureus* (p-values < 0.05). No significant difference was observed for *Pseudomonas aeruginosa*, *Burkholderia cepacia*, *Staphylococcus epidermidis* and *Streptococcus anginosus.* Since these tests were exploratory, no adjustments for multiple testing were made.

Discussion

Summary of findings

The BactInsight system showed the same yield (100%) and similar median TTP as the reference automate (i.e. difference in median TTP of 1.49 h). The median BactInsight TTPs were shorter than the reference for most Enterobacterales, enterococci and some streptococci, and longer than the reference for most non-fermenters, staphylococci, fastidious organisms and yeast. Yield of the turbidimeter on its own was 97.4% and specificity was 85.4%. When comparing the turbidimeter to visual inspection (excluding the fastidious organisms and yeast for which turbidity detection failed), the turbidimeter had an advantage over visual inspection for Enterobacterales, most streptococci and enterococci, but had a longer TTP than visual inspection for *Acinetobacter baumannii* and *Staphylococcus aureus*.

Understanding of the findings

The reference and BactInsight systems have several inherent differences that may have influenced the TTP of (some of) the tested species. The main differences were (i) continuous agitation (for the reference system) versus a static system (for the BactInsight system); a difference in the methodology of growth detection (i.e. colorimetric detection of CO_2 by the reference versus detection of turbidity by the BactInsight system); a difference in measurement frequency (i.e. every 10 min for the reference versus every 30 s for the BactInsight system) and a difference in broth of the blood culture bottles (i.e. not specified complex medium with 0.08% SPS for the reference versus tryptic soy broth with 0.03% SPS for the BactInsight system) and headspace gas (not specified for the reference, not defined for the BactInsight system). Any of these differences may be responsible for (part

Fig. 3. Comparison of the time-to-positivity (TTP, in hours) between the reference system (BACT/ALERT, bioMérieux – boxplot on the left) and the BactInsight system (boxplot on the right). The boxplots show the median TTP, Q1-Q3, ranges and outliers per microorganism and blood culture system and correspond to the data presented in Table 2.

of) the differences in TTP observed between the two systems and may be more or less relevant depending on the tested species. For example, a static system such as the BactInsight system may have disadvantaged those species for which agitation has been shown to decrease the TTP[13].

When compared with manual systems evaluated in the literature, the BactInsight system performed well: two manual blood culture systems evaluated in vitro[12] had a yield of 96% compared to a yield of 100% yield when using the BactInsight system. The two evaluated manual systems showed a day 1 growth (TTP ≤ 24 h) of 75.0% and 90.8%, compared to 89.8% (299/333) for the BactInsight system. Another publication describing the cumulative growth in manual blood cultures in a clinical setting in Cambodia showed an overall day 2 growth of 70.6%[14]. For the BactInsight system tested in vitro, overall growth was 95.2% on day 2 (TTP ≤ 48 h) and 100% on day 3 (TTP ≤ 72 h).

TTP differences and variability of the BactInsight system were largest for fastidious organisms and yeasts (Fig. 3). There were several reasons for this. Firstly, growth detection by the turbidimeter failed for these organisms because of the lack of turbidity during growth. Therefore, we only used the visual inspection results obtained for these organisms. Secondly, some of the runs were started just before the weekend with no reading during the weekend, so that six timepoints for visual inspection were "missed", resulting in an artificial increase in the upper limit of the TTP ranges for these organisms. This happened in 16/28 (57.1%) of the BactInsight BCB sets spiked with fastidious organisms and yeasts, which explains the wide range in TTP found for these organisms. If visual inspection would have been done at regular intervals (as planned), we expect that the BactInsight TTP would have been lower and more comparable to the automate TTP.

When looking at the turbidimeter alone (without taking into account the visual inspection), the yield of the second-generation turbidimeter (BactInsight system) increased significantly compared to the first-generation turbidimeter[10] (Supplementary Table S4). The first-generation turbidimeter had a yield of only 40.0% (24/60, only taking into account growth detected by turbidity), whereas the second-generation turbidimeter showed a yield of 98.5% (128/130) for the reference strains. Of note, for the first-generation turbidimeter, growth detection was assessed using the Growthcurver software in R, simulation experiments were carried out with defibrinated horse blood, and the reference strains tested belonged to only ten different microbial species.

It is important to note that turbidimeter growth detection was based only on the presence of turbidity (i.e. the altered intensity of transmitted and scattered light). This means that species that did not produce turbidity (e.g. fastidious organisms and yeast) or only showed turbidity at a later stage (e.g. *Staphylococcus aureus*) were not or insufficiently detected by the turbidimeter only. In the case of *Staphylococcus aureus*, puff balls at the bottom of the BCB usually appear first, while turbidity follows. In this study, visual growth detection preceded

the turbidimeter in 70.0% of *Staphylococcus aureus* cultures (14/20). This shows the importance of not relying on the turbidimeter alone (in its current version) but using it to complement (and accelerate) visual inspection.

Strengths and limitations

The major strengths of this study were (i) the high number of strains (n = 109, consisting of reference and clinical strains) and microbial species (n = 20) tested, (ii) the standardized simulated test design (based on two publications from Ombelet et al. in 2022[11,12] ensuring the inoculation of appropriate microbial concentrations in the BCBs, and (iii) the use of fresh human blood increasing the comparability with a real-life situation.

Limitations of this study included that (i) only one BACT/ALERT BCB (bioMérieux) was inoculated compared to four BactInsight BCBs, as it was decided to preserve the human blood. In addition, (ii) due to time constraints, some strains (mainly slower growing strains with incubation times up to 96 h) were inoculated on Friday and visually inspected after the weekend (i.e. for *Candida albicans*, *Candida tropicalis*, *Cryptococcus neoformans*, *Haemophilus influenzae*, *Neisseria subflava*, *Burkholderia cepacia*, *Staphylococcus epidermidis* and *Enterobacter cloacae*). For yeast and fastidious organisms, this resulted in an artificial increase in the upper limits of their TTP ranges. Moreover, (iii) due to technical and procedural issues, a substantial amount of data had to be eliminated before data analysis. Additionally, (iv) our study design did not include all required testing to assess the performance of in vitro diagnostic medical devices for regulatory purposes as described in the established guidelines (e.g. CLSI EP05, ISO 20916). This testing is planned in a later phase and will be done on a more advanced version of the turbidimeter prototype.

Implications for future research and conclusion

In a comparative field trial in Benin and Burkina Faso we have evaluated the BactInsight system's performance and acceptability and ease of use in a field setting (unpublished data). In this study, an adapted "tropicalized" turbidimeter (version 2.1) is used in parallel to visual inspection (with discrete measurements done three times daily). In addition, further improvements are being made to the turbidimeter design which will be tested in future field studies in low- and high-resource settings.

In conclusion, the BactInsight system performed well with regard to yield and TTP when compared to the reference system in vitro. The median BactInsight TTPs were shorter than the reference automate in 30.0% of tested species. Fastidious organisms and yeast were challenging to detect by the turbidimeter only and resulted in larger differences in TTP and a higher variability of the BactInsight system when compared to the automate. This study shows that the BactInsight system is a promising system to increase the use of blood cultures in RLS. However, our findings also highlight that the turbidimeter in its current version should complement but not replace visual inspection for growth detection in BCBs.

Data availability

The dataset is available upon request through ITM's contact point for data access (ITMresearchdataaccess@itg.be).

Received: 19 June 2025; Accepted: 14 January 2026
Published online: 22 January 2026

References

1. Ombelet, S. et al. Best Practices of Blood Cultures in Low- and Middle-Income Countries. *Front. Med.* **6**, 131. https://doi.org/10.3389/fmed.2019.00131 (2019).
2. Fernández, L. et al. Developing Diagnostic and Therapeutic Approaches to Bacterial Infections for a New Era: Implications of Globalization. *Antibiotics* **9**(12), 916. https://doi.org/10.3390/antibiotics9120916 (2020).
3. Ventura, F., Greub, G., Liles, W. C. & Jacob, S. T. Proposed Framework for Conducting Clinically Relevant Translational Biomarker Research for the Diagnosis, Prognosis and Management of Sepsis. *Diagnostics* **14**(3), 300. https://doi.org/10.3390/diagnostics14030300 (2024).
4. World Health Organization. Global action plan on antimicrobial resistance. (2015). https://www.who.int/publications/i/item/9789241509763
5. Lamy, B., Dargère, S., Arendrup, M. C., Parienti, J. J. & Tattevin, P. How to optimize the use of blood cultures for the diagnosis of bloodstream infections? A State-of-the Art. *Front Microbiol* **7**, 697. https://doi.org/10.3389/fmicb.2016.00697 (2016).
6. Fabre, V., Carroll, K. & Cosgrove, S. Blood culture utilization in the hospital setting: a call for diagnostic stewardship. *J. Clin. Microbiol.* **60**, e01005–e01021. https://doi.org/10.1128/JCM.01005-21 (2022).
7. Ondoa, P. et al. Bacteriology testing and antimicrobial resistance detection capacity of national tiered laboratory networks in sub-Saharan africa: an analysis from 14 countries. *Lancet Microbe* **6**, 100976. https://doi.org/10.1016/j.lanmic.2024.100976 (2025).
8. Jacobs, J. et al. Diagnostic bacteriology in district hospitals in sub-Saharan africa: at the forefront of the containment of antimicrobial resistance. *Front Med. (Lausanne)* **6**, 205. https://doi.org/10.3389/fmed.2019.00205 (2019).
9. Dailey, P. J. et al. Defining System Requirements for Simplified Blood Culture to Enable Widespread Use in Resource-Limited Settings. *Diagnostics* **9**(1), 10. https://doi.org/10.3390/diagnostics9010010 (2019).
10. Barbé, B. et al. Pilot Testing of the Turbidimeter, a Simple, Universal Reader Intended to Complement and Enhance Bacterial Growth Detection in Manual Blood Culture Systems in Low-Resource Settings. *Diagnostics* **12**(3), 615. https://doi.org/10.3390/diagnostics12030615 (2022).
11. Ombelet, S. et al. Considerations in evaluating equipment-free blood culture bottles: A short protocol for use in low-resource settings. *PLoS One.* **17**(4), e0267491. https://doi.org/10.1371/journal.pone.0267491 (2022).
12. Ombelet, S. et al. Biphasic versus monophasic manual blood culture bottles for low-resource settings: an in-vitro study. *Lancet Microbe.* **3**(2), e124–e132. https://doi.org/10.1016/S2666-5247(21)00241-X (2022).
13. Hawkins, B. L. & Peterson, E. M. Maza, L. M. Improvement of positive blood culture detection by agitation. *Diagn. Microbiol. Infect. Dis.* **5**, 207–213 (1986).
14. Peeters, M. et al. Slow growth of *Burkholderia pseudomallei* compared to other pathogens in an adapted blood culture system in Phnom Penh, Cambodia. *J. Med. Microbiol.* **68**(8), 1159–1166. https://doi.org/10.1099/jmm.0.001011 (2019).

Acknowledgements
We want to thank all volunteers who have donated blood for the study. We would also like to thank our partners with whom we collaborate on the blood culture surveillance who provided clinical isolates for this study.

Author contributions
Conceptualization, L.H. ; methodology, B.B, E.C. and L.H. ; software J.C., M.G. and F.M. ; validation Y.L., R.B., J.J and L.H. ; formal analysis E.G., B.B., J.C. and L.H. ; writing – original draft preparation B.B. and J.C. ; writing – review and editing: M.G., E.C., E.G., F.M., Y.L., R.B., J.J., L.H.; visualisation B.B., J.C., E.G. and L.H. ; supervision L.H. ; funding acquisition: L.H. All authors have read and agreed to the published version of the manuscript.

Funding
This study is part of the EDCTP2 programme (grant number RIA2020I-3270-SIMBLE) supported by the European Union.

Declarations

Competing interests
The authors declare no competing interests.

Additional information
Supplementary Information The online version contains supplementary material available at https://doi.org/10.1038/s41598-026-36676-y.

Correspondence and requests for materials should be addressed to B.B.

Reprints and permissions information is available at www.nature.com/reprints.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.