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Evaluation of an ID Agnostic eQUANT™ RUO System for Use on Gram-Negative Bacillus and Yeast

I. Introduction

The eQUANT™ system is a compact benchtop sample preparation device that utilizes proprietary electrochemical biosensor technology to quantify microorganisms in suspension by measuring changes in oxidation-reduction potential (ORP) associated with microbial growth. In its current FDA-cleared IVD configuration, eQUANT™ requires prior identification of the Gram-negative organism recovered from positive blood culture in order to generate a standardized bacterial inoculum equivalent to a 0.5 McFarland (eMcFarland), which serves as input for downstream disk diffusion antibiotic susceptibility testing (AST).

Here, we present proof-of-concept data for a next-generation research use only (RUO) eQUANT™ system that does not require prior species identification to generate a standardized bacterial suspension within a target concentration range of 1–2 × 10^8 CFU/mL (±0.6 log).

In contrast to the current IVD eQUANT™ system, which aerates only aerobic organism samples and applies species-specific algorithms to generate a 0.5 McFarland equivalent suspension, this next-generation system utilizes aeration across all organism types and applies a universal algorithm for generation of a standardized output sample.

By developing the ID agnostic eQUANT™ RUO system, we aim to not only simplify its workflow but also to expand controlled microorganism growth capabilities for a broader range of downstream applications beyond antibiotic susceptibility testing, including MALDI-TOF ID.

II. Method

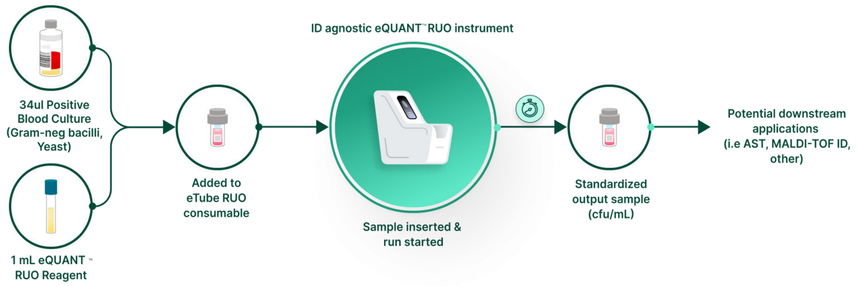

Figure 1. ID agnostic eQUANT RUO workflow. To start a run, 34ul of positive blood culture and 1mL of eQUANT reagent are transferred into the eTube consumable. After a brief vortex step, the sample is inserted into the instrument and the run is started. After run completion, the instrument cools the output sample to 15°C to inhibit growth and can hold it for up to 1 hour.

Clinically significant Gram-negative bacilli (GNBs) and yeast were grown up from a frozen stock prior to being spiked into Biomérieux Standard Aerobic blood culture bottles which were incubated to positivity and processed on the eQUANT™ RUO System. The isolates tested from contrived positive blood culture bottles were *A. baumanii* (5), *A. Iwoffii* (1), *A. pittii* (1), *A. xylosoxidans* (6), *C. freundii* (5), *E. coli* (5), *E. cloacae* (5), *K. aerogenes* (5), *K. oxytoca* (5), *K. pneumoniae* (5), *M. morganii* (11), *P. mirabilis* (5), *P. vulgaris* (5), *P. aeruginosa* (5), *P. putida* (1), *S. marcescens* (5), *S. maltophilia* (5), *C. auris* (1) and *C. tropicalis* (2) and were procured from CDC, ATCC, Stanford Clinical Microbiology Laboratory, UCLA Clinical Microbiology, University Hospitals Clevland Medical Center, BEI Resources, International Health Management, Associates, and John Muir Clinical Microbiology Laboratory. Positive blood cultures (PBCs) were processed on the eQUANT™ RUO instrument within 12 hours of the blood culture flagging positive. PBCs were plated for colony counts on blood agar plates prior to eQUANT™ setup. For all eighty-eight (88) samples, 1 mL of the eQUANT™ reagent (CAMHB with 0.0015% antifoam) and 34 µL of PBC were transferred into the eTube consumable. The eTube was vortexed before loading onto the eQUANT™ RUO System (see Figure 1). Upon run completion, the run time was recorded, and the output sample was processed for colony counts on blood agar within 10 minutes following removal from the instrument to evaluate if the target concentration was achieved.

III. Results

The system performance was evaluated two-fold: 1) for the run times required to generate the organism output from Gram-negative bacilli and yeast PBCs and 2) for meeting the target concentration of 1-2 e^8 CFU/ml +/- 0.6 log (2.51xe^7 - 7.96xe^8 CFU/mL) by performing colony counts of the eQUANT™ output sample.

1) Run times

A total of 88 samples were run on the ID agnostic eQUANT™ RUO system and completed in an overall average of 110 minutes (Figure 2, Table 1). Most Enterobacterales samples completed in an average of 66 minutes, whereas some aerobic species including *Acinetobacter lwoffii* and *Pseudomonas aeruginosa* required more time and completed in 2 hours on average (Table 1). As shown in Table 1 and Figure 1, four organisms demonstrated slower growth times and included *Achromobacter xylosoxidans* (340 minutes average run time), *Stenotrophomonas maltophilia* (175 minutes average run time), and *Candida* species *(Candida auris* and *tropicalis*, 329 minutes average run time). Varying run times were attributed to different starting concentrations due to a wide range of organism concentrations in positive blood cultures (see Figure 3) and different organism growth rates.

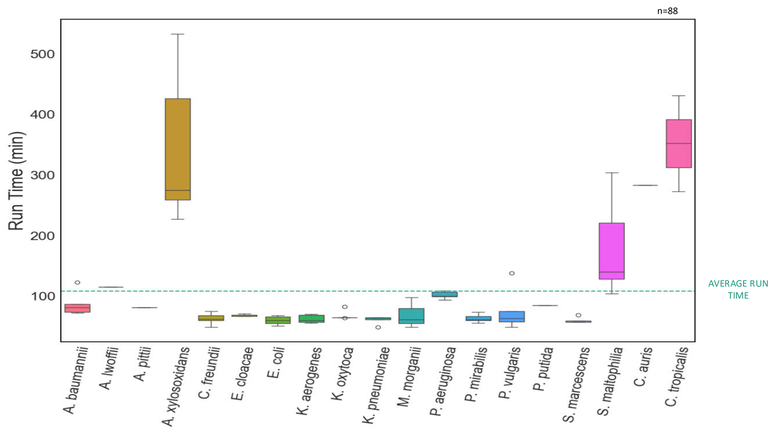

Figure 2. Average Sample Run Times. After each run completed, run times were recorded and plotted by organism type. The overall run time average was 110 minutes.

Organism	Isolates (n)	Average Run Time (min)
Acinetobacter species	7	90
Achromobacter xylosoxidans	6	340
Candida species	3	329
Enterobacterales	56	66
Pseudomonas species	6	99
Stenotrophomonas maltophilia	10	175
Overall	**88**	**110**

Table 1. Average Sample Run Times.

2) Output sample concentration range

Positive Blood Cultures are the eQUANT input samples. As previously described (Abdelhamed et al., 2025; Dutta et al., 2025) and demonstrated here, blood cultures positive for Gram-negative bacilli and yeast, span a wide concentration range between 1e^8 to 1.96e^{10} CFU/mL upon positivity (Figure 3). As shown in Figure 4, our next-generation ID agnostic eQUANT™ RUO system successfully generated standardized organism outputs for all 88 samples tested and in the target range of 1-2 e^8 CFU/ml +/- 0.6 log (2.51xe^7 - 7.96xe^8 CFU/mL), despite a >2log range in PBC starting concentrations. This indicates that the eQUANT™ RUO System in its new configuration can consistently produce a standardized inoculum for Gram-negative bacilli and yeast PBCs without requiring prior organism identification.

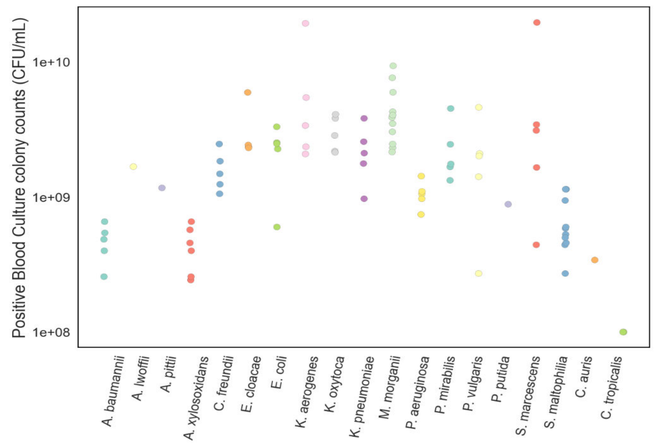

Figure 3. Positive Blood Cultures concentration range.

Figure 4. Concentrations of output samples after run completion.

IV. Conclusions and Outlook

In this proof-of-concept, the Avails RUO ID agnostic version of the eQUANT™ system demonstrates the ability to generate a standardized organism sample of 1-2 e^8 CFU/ml +/-0.6 log directly from positive blood cultures containing Gram-negative bacilli AND yeast without prior organism identification.

With an output concentration equivalent to the eMcFarland produced by the eQUANT™ IVD version, eQUANT™ RUO has the potential to be used for downstream AST and other applications including MALDI-TOF ID. It also presents the potential to tailor the output concentration to specific downstream applications by algorithm adjustments or accelerate run times through reagent optimization.