# Evaluation of Qvella's FAST-Prep™ Liquid Colony™ for Early Antimicrobial Sensitivity Testing of Positive Blood Culture by Disk Diffusion Method

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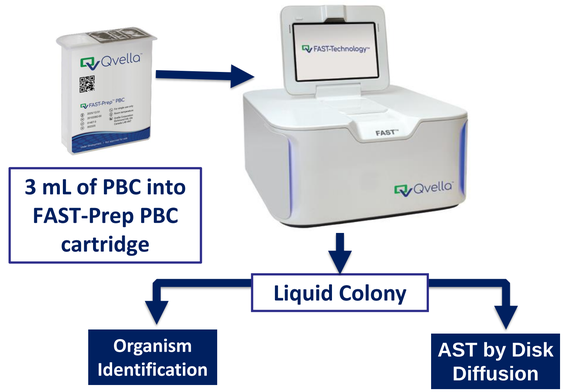
## BACKGROUND

Conventional antimicrobial susceptibility testing (AST) of microorganisms from positive blood cultures (PBC) can take ≥ 2 days. In order to improve the turnaround time for AST on a PBC, CLSI and EUCAST have made efforts to standardize procedures for disk diffusion (DD) direct from a PBC. Qvella (Richmond Hill, ON, CA) has developed the FAST-Prep™ System, an automated centrifugal sample preparation system that rapidly delivers a Liquid Colony: a clean, concentrated, viable pathogen cell suspension directly from a PBC. This study was performed to investigate the feasibility of DD AST using a Liquid Colony.

## OBJECTIVES

- Compare the biomass from a Liquid Colony with the biomass in a positive blood culture.
- Compare the biomass in 0.5 McFarland derived from Liquid Colony with a 0.5 McFarland derived from a solid colony.
- Verify that a 0.5 McFarland suspension, from a Liquid Colony can be used as the inoculum for conventional DD testing (using CLSI M-100 interpretative criteria).

## FAST-Prep WORKFLOW




*Gram Stain and subculture performed on all PBC*

The FAST-Prep System is under development and not approved for sale. The performance characteristics have not been established

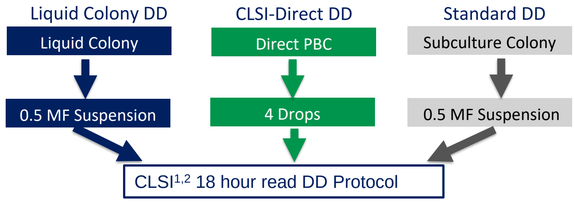
## METHOD

### Contrived PBC Preparation and Processing


### Biomass Comparison


### Biomass Comparison of 0.5 McFarland (MF) Suspension


### Disk Diffusion (DD) AST




| Gram-Positive Bacteria Susceptibility Panels | | |
|---|---|---|
| *Enterococcus* spp. | *Staphylococcus* spp. | *Streptococcus* spp. |
| Ampicillin | Cefoxitin | Clindamycin |
| Ciprofloxacin | Clindamycin | Erythromycin |
| Erythromycin | Erythromycin | Levofloxacin |
| Levofloxacin | Levofloxacin | SXT |
| Tetracycline | Tetracycline | |
| Vancomycin | SXT | |

| Gram-Negative Bacteria Susceptibility Panels | | |
|---|---|---|
| *Enterobacteriaceae* | *P. aeruginosa* | *Haemophilus* spp. |
| *Acinetobacter* spp. | | |
| Amikacin | Amikacin | Ampicillin |
| Ceftazidime | Ceftazidime | Ciprofloxacin |
| Ciprofloxacin | Ciprofloxacin | Levofloxacin |
| Meropenem | Levofloxacin | SXT |
| Piperacillin/Tazobactam | Meropenem | |
| SXT | Piperacillin/Tazobactam | |

## RESULTS

Table 1. Average Biomass in PBC and Liquid Colony

| Test Sample | | CFU/mL | |
|---|---|---|---|
| Species | Strain (n) | PBC | Liquid Colony |
| *Enterococcus faecalis* | 5 | 2.87 x $10^9$ | 1.11 x $10^9$ |
| *Enterococcus faecium* | 5 | 4.84 x $10^8$ | 2.04 x $10^9$ |
| *Staphylococcus aureus* | 5 | 2.83 x $10^7$ | 7.79 x $10^7$ |
| *Staphylococcus lugdunensis* | 4 | 5.54 x $10^7$ | 3.07 x $10^8$ |
| *Streptococcus pneumoniae* | 4 | 4.54 x $10^8$ | 4.99 x $10^8$ |
| *Streptococcus pyogenes* | 3 | 9.85 x $10^8$ | 2.89 x $10^8$ |
| **Gram-positive average** | | **8.13 x $10^8$** | **7.20 x $10^8$** |
| *Escherichia coli* | 3 | 1.78 x $10^9$ | 4.03 x $10^9$ |
| *Acinetobacter baumannii* | 3 | 3.02 x $10^8$ | 4.82 x $10^8$ |
| *Pseudomonas aeruginosa* | 4 | 4.52 x $10^8$ | 3.72 x $10^8$ |
| *Haemophilus influenzae* | 3 | 1.87 x $10^9$ | 5.64 x $10^8$ |
| **Gram-negative average** | | **1.04 x $10^9$** | **1.23 x $10^9$** |

Table 2. Average Concentrations in Standardized 0.5 MF Suspensions

| Test Sample | | CFU/mL | |
|---|---|---|---|
| Species | Strain (n) | Subculture | Liquid Colony |
| *Enterococcus faecalis* | 5 | 4.78 x $10^7$ | 6.82 x $10^7$ |
| *Enterococcus faecium* | 5 | 7.66 x $10^7$ | 5.38 x $10^7$ |
| *Staphylococcus aureus* | 5 | 7.44 x $10^7$ | 3.08 x $10^6$ |
| *Staphylococcus lugdunensis* | 4 | 9.80 x $10^7$ | 1.42 x $10^7$ |
| *Streptococcus pneumoniae* | 4 | 5.78 x $10^7$ | 6.80 x $10^7$ |
| *Streptococcus pyogenes* | 3 | 5.77 x $10^7$ | 1.95 x $10^7$ |
| **Gram-positive average** | | **6.87 x $10^7$** | **3.78 x $10^7$** |
| *Escherichia coli* | 3 | 9.27 x $10^7$ | 7.60 x $10^7$ |
| *Acinetobacter baumannii* | 3 | 6.80 x $10^7$ | 3.43 x $10^7$ |
| *Pseudomonas aeruginosa* | 4 | 2.50 x $10^8$ | 5.60 x $10^7$ |
| *Haemophilus influenzae* | 3 | 9.80 x $10^7$ | 9.47 x $10^7$ |
| **Gram-negative average** | | **1.15 x $10^8$** | **5.98 x $10^7$** |

Table 3. Disk Diffusion Antimicrobial Susceptibility Test Comparison

CA; categorical agreement, mE; minor error, ME; major error

| Test Sample | | DD from FAST-Prep Liquid Colony | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Category | Strain (n) | Test (n) | CA | S | I | R | mE | ME |
| Gram-positive bacteria | 25 | 136 | 130 | 80 | 19 | 33 | 6 | 0 |
| % | | | **95.6** | | | | **4.4** | **0.0** |
| Gram-negative bacteria | 13 | 72 | 71 | 49 | 11 | 12 | 1 | 0 |
| % | | | **98.6** | | | | **1.4** | **0.0** |

| Test Sample | | CLSI-Direct DD | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Category | Strain (n) | Test (n) | CA | S | I | R | mE | ME |
| Gram-positive bacteria | 25 | 136 | 105 | 62 | 22 | 48 | 28 | 3 |
| % | | | 77.2 | | | | 20.6 | 4.8 |
| Gram-negative bacteria | 13 | 72 | 59 | 38 | 20 | 13 | 12 | 1 |
| % | | | 81.9 | | | | 16.7 | 2.6 |

## CONCLUSIONS

- Biomass obtained from Liquid Colony is sufficient to prepare a 0.5 McFarland suspension for DD susceptibility testing.
- DD AST using a Liquid Colony resulted in a CA of ≥95% and improved performance than CLSI-Direct DD when compared to standard DD AST resulting in no ME and VME.
- FAST-Prep PBC generated Liquid Colony can be potentially used to set up an AST 24 hours earlier than a conventional subculture colony.

## REFERENCES

[1]CLSI. 2018. Performance standards for antimicrobial disk susceptibility tests. 13th ed. CLSI supplement M02. Clinical and Laboratory Standards Institute, Wayne, PA.
[2]CLSI. 2019. Performance standards for antimicrobial susceptibility testing. 29th ed. CLSI supplement M100. Clinical and Laboratory Standards Institute, Wayne, PA.
[3]Chandrasekaran, S., et. al. 2018. Direct-from-blood-culture disk diffusion to determine antimicrobial susceptibility of gram-negative bacteria: preliminary report from the Clinical and Laboratory Standards Institute methods development and standardization working group. J. Clin. Microbiol. 56:1-10.