Autosampler-Ready Microporous Filter Vials As A Viable Alternative For Microbiological Sample Processing

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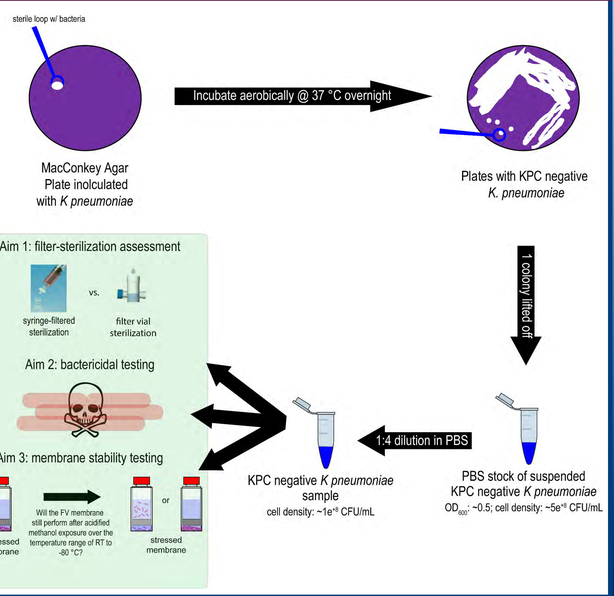
Introduction

Demand for high-throughput sample processing technologies will increase as LC-based methods become more prevalent in academic and clinical microbiological research labs[1,2]. Filter sterilization steps are required during the processing of microbiological samples because of the following[3]: i) many bacteria and fungi are BSL 2/3 level pathogens; ii) used broth media tend to contain solids; and, iii) chromatographic column longevity is required in a high-throughput laboratory[4].

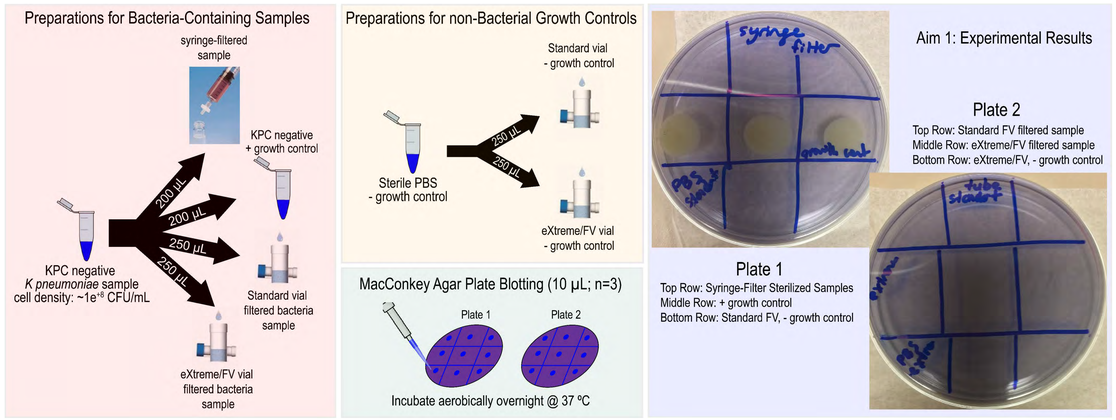
Project Aims

1. Perform a side-by-side comparison between Thomson Standard and eXtreme/FV® autosampler-ready microporous filter vials (0.2 micron) and a typical syringe-based filtration device (0.22 micron)[5];
2. Identify a bactericidal solvent system that is capable of killing the suspended gram negative bacteria *Klebsiella pneumoniae*;
3. Assess the stability the filter vial membrane while submersed for 6 days in a non-bactericidal solvent system (25:75 methanol:water + 0.1% acetic acid) at RT, +4 °C, -20 °C, and -80 °C.

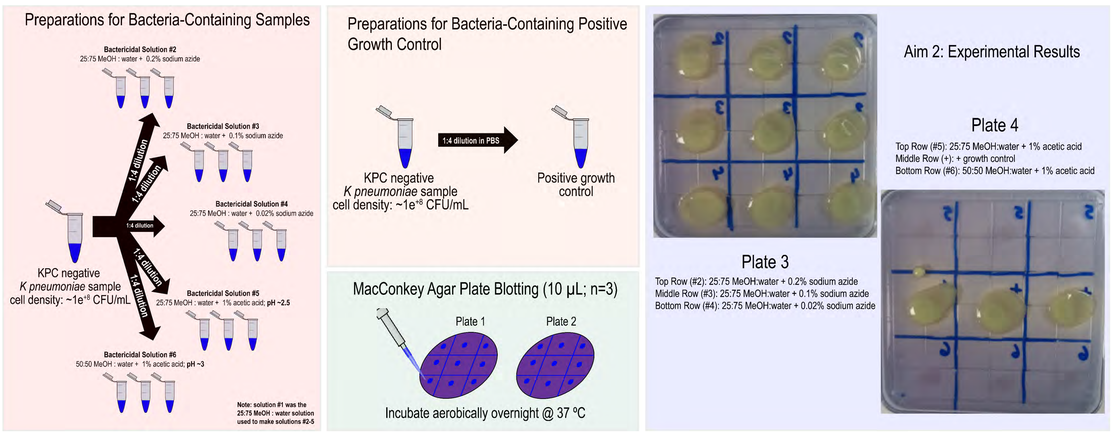
Bacterial Expansion and Study Design



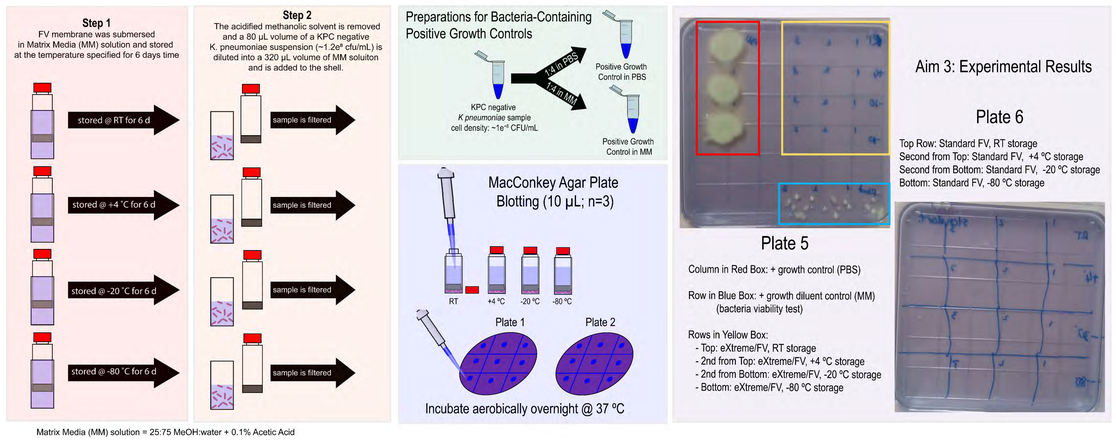
Aim 1 Experiment / Results



Aim 2 Experiment / Results



Aim 3 Experiment / Results



Matrix Media (MM) solution = 25:75 MeOH:water + 0.1% Acetic Acid

Observations

- Aim 1: Filter vials are as effective at eliminating suspended gram negative bacteria *K. pneumoniae* from a media sample as a gold standard syringe-based filter system.
- Aim 2: Solutions consisting of methanol, water, and 1% acidic acid proved to be bactericidal to *K. pneumoniae* – a pH below 4 seems to drive the effect.
- Aim 3: After six days of submersion storage in a MM solution (25:75 methanol:water + 0.1% acetic acid) at RT, +4 °C, -20 °C, and -80 °C, the filter vial membranes remained intact and effective at filter-sterilizing *K. pneumoniae* bacteria suspended a MM solution.

Conclusions

The Thomson Standard and eXtreme/FV® filter vials have been shown to be as effective at eliminating *K. pneumoniae* from a sample as a standard syringe-based filter system. The integrated filter membrane has been shown to remain stable under long-term storage (six days) in an acidified methanolic solution at temperatures that range from RT to -80 °C.

Future Work

- Adapt the usage of the Thomson filter vials for our in-house LC-MS/MS-based method for assessing isolate-specific antimicrobial resistance (AMR) patterns in *K. pneumoniae.*
- We assess the performance of the Thomson filter vials for filter sterilization of suspended gram positive bacteria, such as *Staphylococcus aureus*.

Acknowledgements

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