

# Shape-based Separation of Mastitis Pathogens Using Nanosculpted Membranes

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## Introduction

A 2019 report from UW-Madison shows that the Wisconsin dairy industry accounts for over half of the total annual revenue from Wisconsin agriculture.<sup>1</sup> Diseases in dairy cattle threaten profit and production with mastitis alone causing an estimate annual loss of \$1B nationally and \$140M locally.<sup>2</sup> Broad spectrum antibiotics, the current regimen for mastitis has the potential to generate antibiotic resistant pathogens. The ability to isolate the cause of individual mastitis cases will lead to tailored treatment plans resulting in lower costs, shorter lost production windows, and less risk of future pathologic issues. This project seeks to exploit the distinct aspect ratios found among different pathogens by fabricating customized nanosculpted membranes. In the future, dairy farms can use an onsite testing device to identify the source of infection and implement a treatment plan.

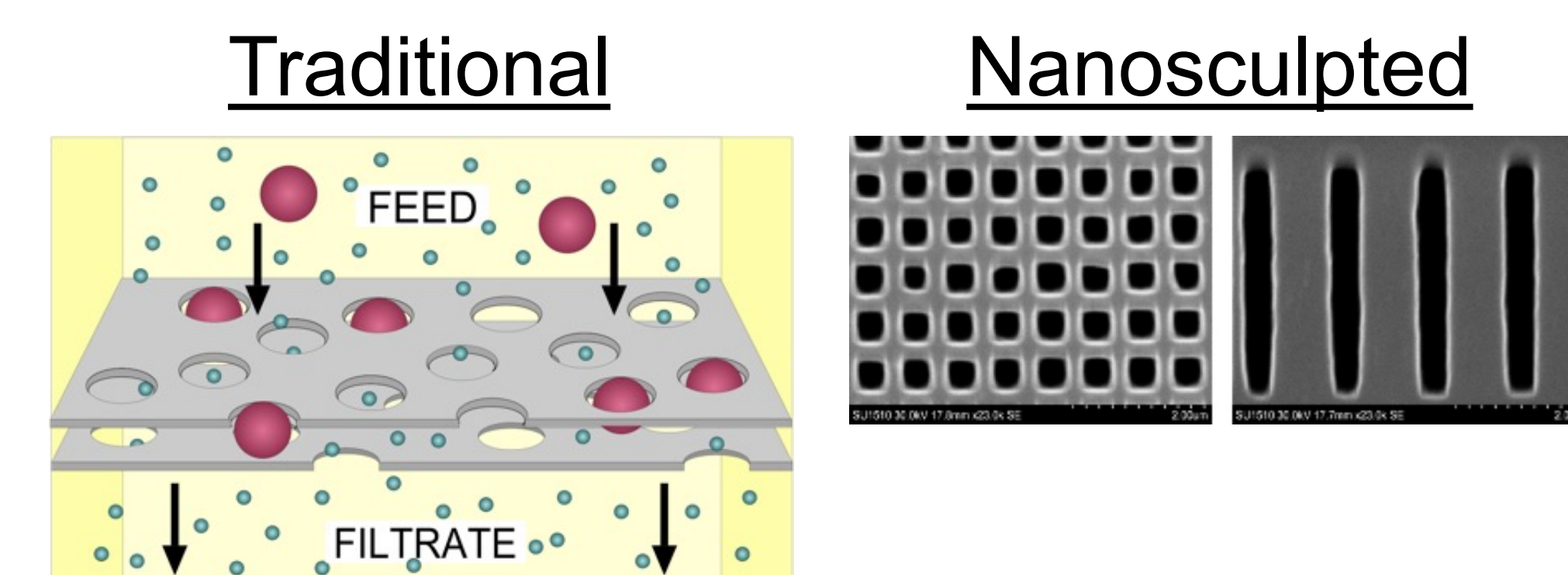
## Mastitis

- Most common dairy cattle disease
- Caused by ~20 known pathogens
- Treated with broad spectrum antibiotics
- Antibiotic resistance is a serious threat



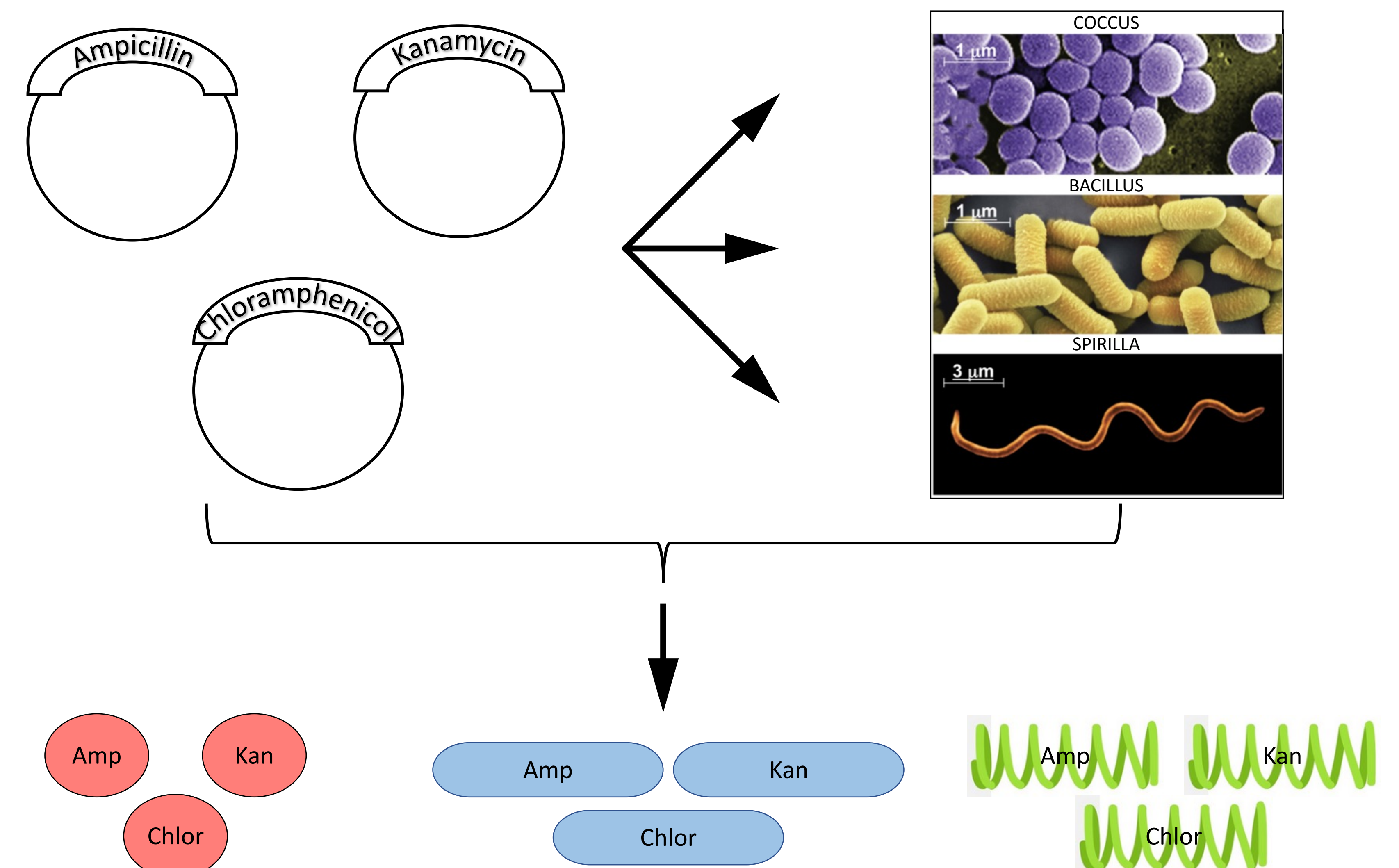
## Nanosculpted Membrane

- Traditional separations use circular pores based on average mass
- Many objects have similar masses but disparate aspect ratios
- “Nanosculpting” generates custom pore patterns at nanometer tolerances

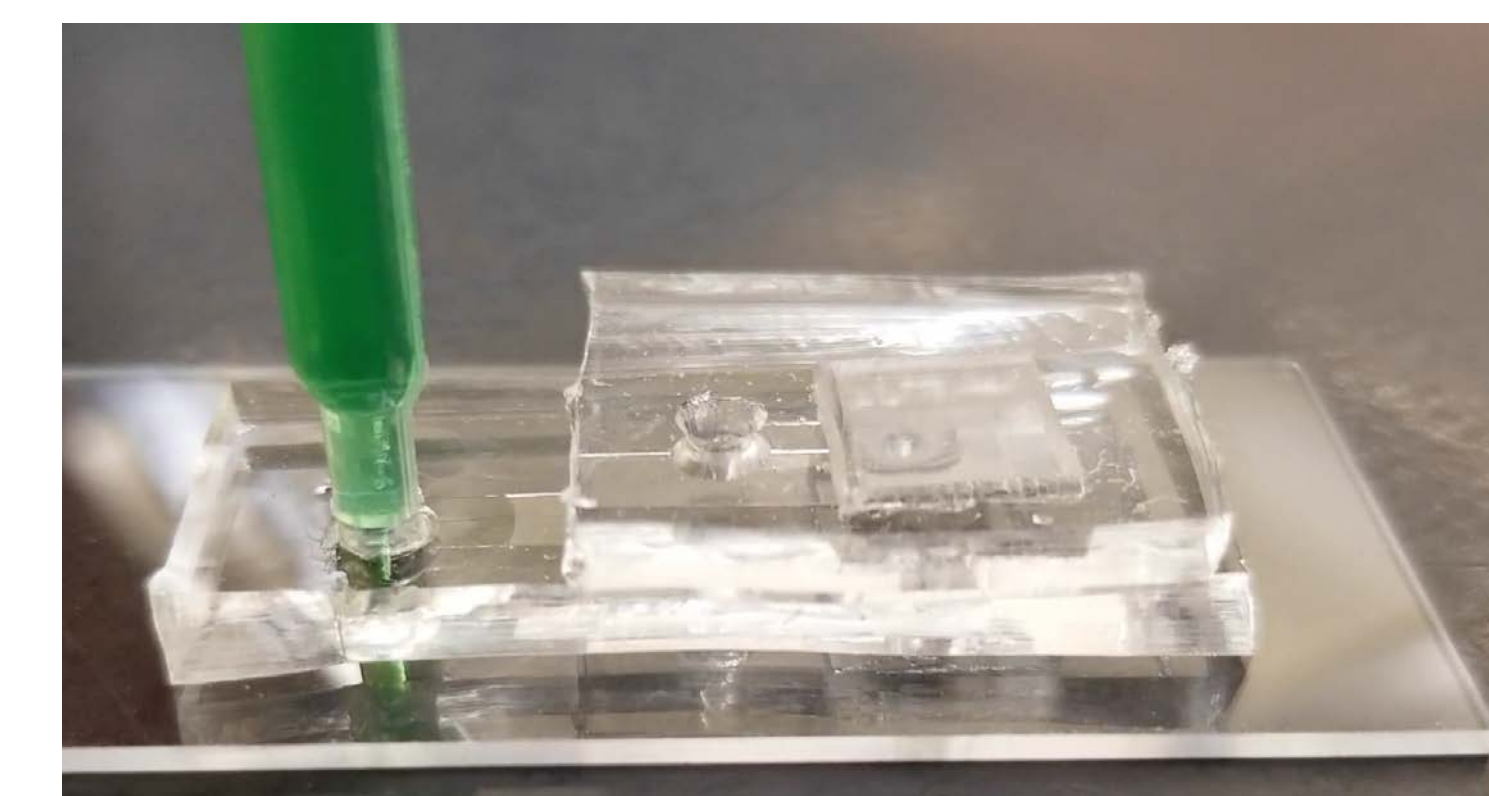


## Approach

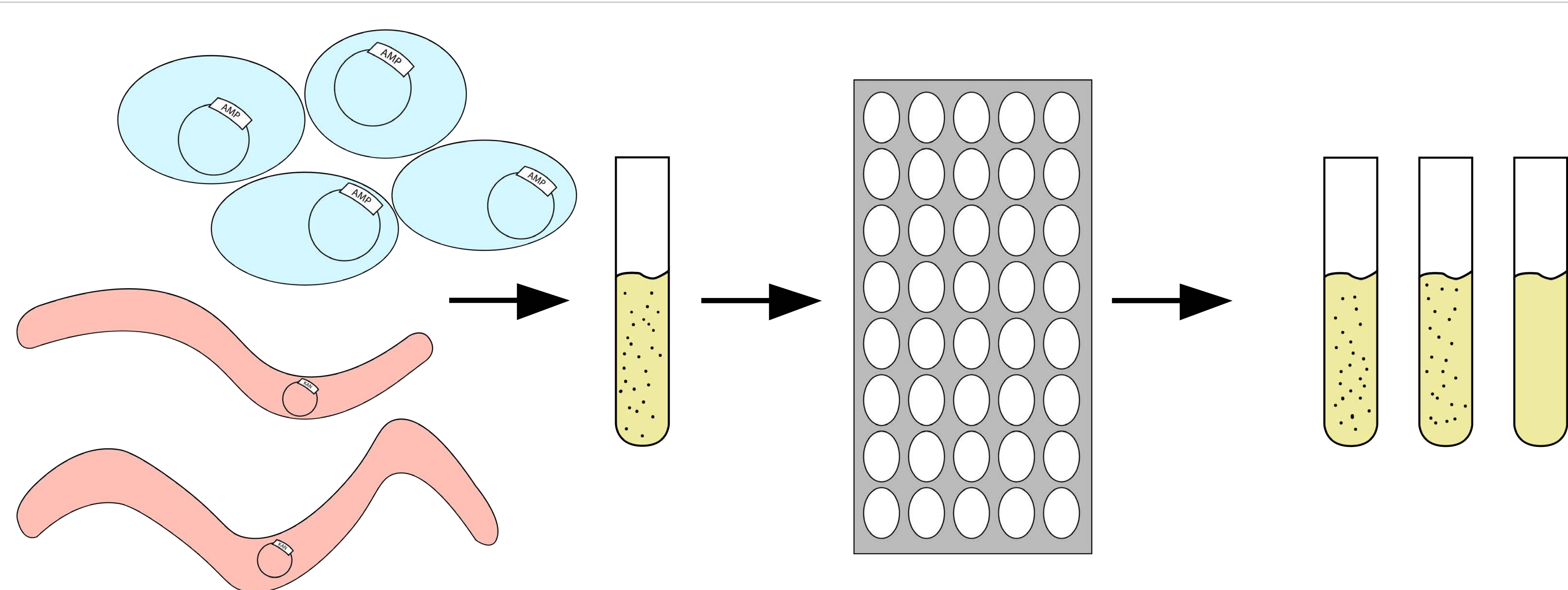
### Library Generation



### Nanomembrane Prototype



## Assay



- Complex solution prepared containing multiple bacteria species with distinct antibiotic resistance markers
- Mixture applied to nanosculpted membrane customized to a target aspect ratio
- Filtrate transferred to an array of test media containing different antibiotics
- Target bacteria identified by successful growth in selective media

## Accomplishments and Future Directions

- Generated a collection of unique bacteria samples
  - Distinct aspect ratios
  - Different antibiotic resistance markers
- Multiple approaches to develop reagents
  - Electroporation of plasmid:bacteria combinations
  - Heat Shock transformation of chemically competent bacteria
- Created a nanosculpted filtration prototype
  - Currently assaying filter tolerances with mixed sample cultures
- Goal – portable test filter to identify specific cause of infection
  - Suitable for on-site testing by dairy farmers

### References:

- <sup>1</sup>Deller, S. 2019. The Contribution of Agriculture to the Wisconsin Economy: An Update for 2017.  
<sup>2</sup>Ruegg, P. 2011. Premiums, Production and Pails of Discarded Milk How Much Money Does Mastitis Cost You?