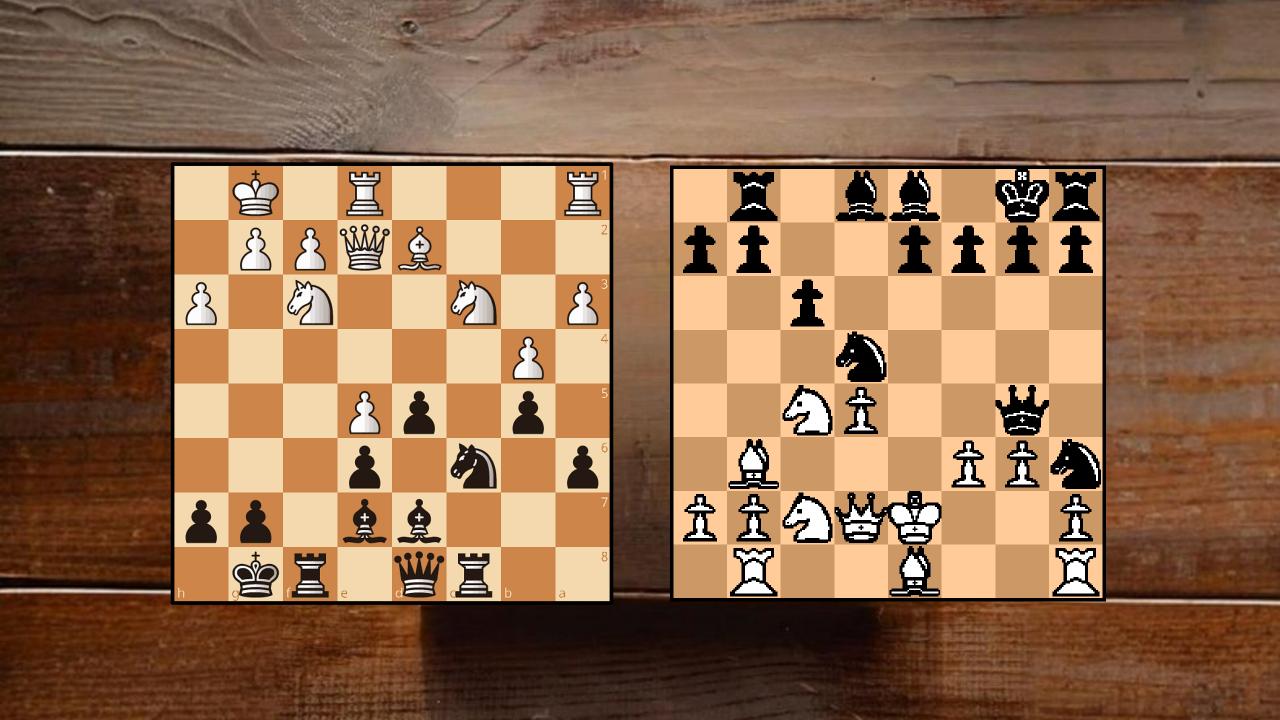


Sustainability in Science

Integrating It Safely and Efficiently

By Patrick Penndorf



How My Journey Began







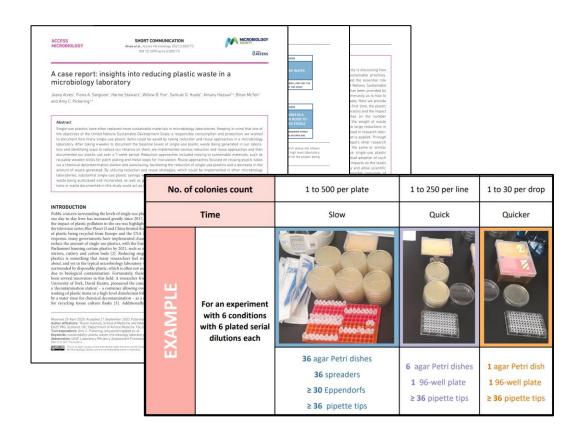
35-65% Less Plastic Waste in Sterile Conditions

It Is Possible – And Has Been Done





35-65% Less Plastic Waste in Sterile Conditions



Saving 516kg plastic >\$1500

It Is Possible – And Has Been Done

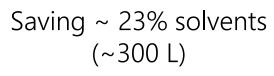














Saving 2 Mio liters of Nitrogen

Saving 31.8 Million kWh

What Your Colleagues Have Done



A List of Sustainable Practices

Reducing waste

Reducing plastic waste within the laboratory can be achieved by:

- Optimizing protocols to prepare solutions in a single instead of separate tubes
- Using items in a smart manner by rethinking their application possibilities (e.g., mixing loading dye and samples for agarose gels after kit use on a piece of parafilm or within a reusable mixing plate instead of separate tubes)
- Using alternatives such as glass or metal items for flasks, dishes, and serological pipettes)
- Minimizing the size of consumables (especially tubes, serological pipettes, pipette tips to the required volume, potentially pipetting twice)
- Optimize for the least necessary conduct (e.g., leaving out parafilm when evaporation of fluid is not an issue)
- Pouring solutions where precises volumes are not decisive (e.g., washing steps)
- Reusing Falcon tubes, potentially after rinsing, especially for frequently used solutions (e.g., Tris solutions when preparing SDS-PAGE nels)
- Reusing pipette tips, tubes where cross-contamination is not an issue (e.g., pipetting samples in agarose gels to control for restriction digests)
- Reusing items (e.g., electroporation cuvettes or tissue/cell strainers after cleaning, cuvettes for OD measurements, weighting boards)
- Precise calculation and bulk preparation of reagents and solutions
- Automatization and miniaturization through automated systems which pipette more precisely and are less error prone
- Conscious use of gloves (see our <u>previous lesson</u> to learn all you need to know)
- Using items to their capacity (e.g., maximize use of wells in a 96-well plate, all targets on an MS plate or both ends of a toothpick to pick colonies instead of pipette tips)
- Sharing items and resources to their maximum potential (e.g., parts of unused agarose gels can be kept in buffer for up to 1

- Making use of take-back programs for plastic items, including Styrofoam
- Separating waste into plastic, paper, others, autoclavation (e.g., plastic wrapping from serological pipettes tips can be discarded in plastic and paper after separating instead of

Improving experimental conduct and design

- Proper experimental planning can be achieved by:
 - Leveraging existing literature to avoid redundant experiments
 - Robust statistical planning (especially power analysis to assure interpretable outcomes) help reduce sample sizes and enhance statistical validity (e.g., group sequential design for work entailing animals)
 - Carefully chosen experimental conditions with proper controls (e.g., using painkillers with known mechanism of actions for control conditions)
 - Reducing use of animals by switching to in vitro / in silico or smart experimental design (e.g., reducing breeding through smart knock out/in strategies)
 - Re-optimizing protocols that were tailored for a specific tissue/sample/chemical
 - Reviewing possibilities for optimization in consumable utilization ahead of conduct (including material, size and number of consumables)
 - Exploring preparation procedures (e.g., optimizing pipetting schemes and master-mixes to reuse tips and tubes)
- Optimization of experimental produces is possible through:
 - Adopting safer and more benign alternatives for commonly used reagents in experiments (e.g., DNA staining solutions, microscopic slide mounting agents, lysing agents, or protease inhibitors)
 - Exploring alternative experimental approaches (e.g., using Supercritical fluid chromatography to avoid organic solvents needed in HPLC or modes of inducing cell death)

- Minimization of experimentation (e.g., reducing PCR volumes to 10uL instead of 20uL if the overall presence of a construct or the success of a digestion should be assessed)
- Implementing strategies and frameworks to ensure best practices (e.g., handling pipettes upright when pipetting)
- Awareness of toxicity of reagents in use for proper handling and discarding (e.g., including closing lids to avoid evaporation)
- · Impact can be reduced by initiating collaboration with:
- Colleagues in co-preparation of solutions, sharing of samples or co-use of machines (e.g., water baths)
- Other groups to share equipment
- Core facilities or partners to avoid unnecessary establishment of new methods
- -> Cool Example from my consulting:

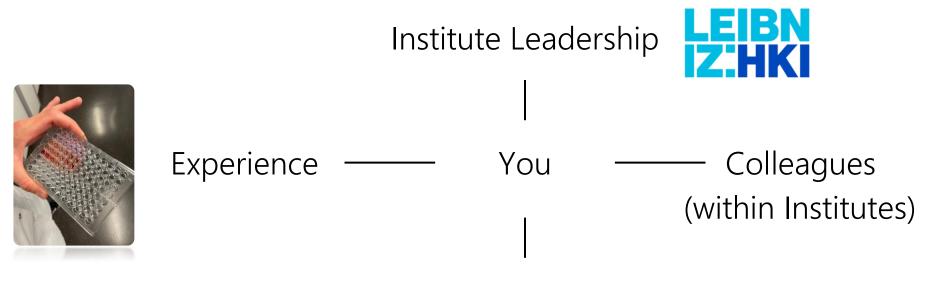
One of the scientists decided to use larger 2 mL tubes instead of 1.5 mL tubes. The reason: since the 2 mL ones have a round bottom instead of the conical shape of the 1.5 mL tubes, he could resuspend the pellet by vortexing without needing a pipette. Additionally, he could pour off the supernatant after centrifugation without needing to pipette it out, since the larger surface ensured the pellet adhered to the bottom. In doing so, he saved two tips and quite a bit of time.

Changing procurement and purchasing processes

- · Planning orders carefully by:
 - Creating an internal system to track chemical inventory and consumable supplies to minimize unnecessary orders
 - Reduce frequency of orders
 - Collaborating with other laboratories or facilities to collect orders

You Have All The Support





Next Generation

1st Case Studies



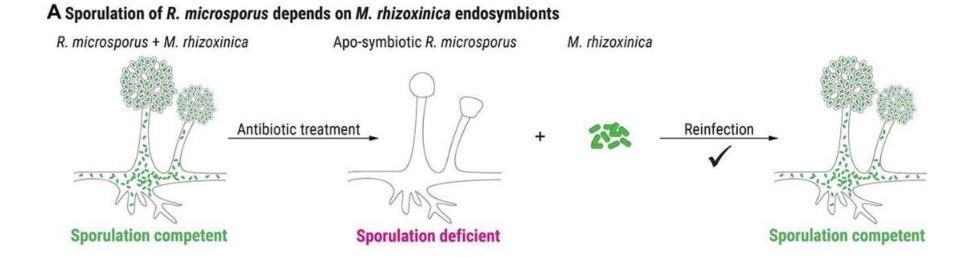
Biomolecular Chemistry - Dr. Jana Krabbe & Dr. Evelyn Molloy

- Understanding Experimental Processes
- Creating A Lab-Culture
- Considering Individual Preferences

1st Case Study



Biomolecular Chemistry - Dr. Jana Krabbe & Dr. Evelyn Molloy



DOI: 10.1093/ismejo/wrae074

1st Case Study



Biomolecular Chemistry - Dr. Jana Krabbe & Dr. Evelyn Molloy







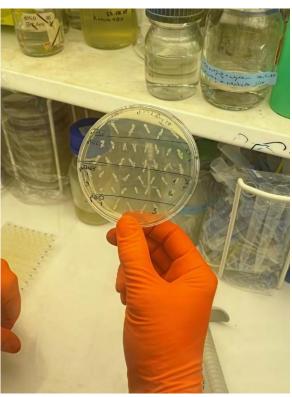
Holding Temp. 12-16°C

1st Case Study



Biomolecular Chemistry - Dr. Jana Krabbe & Dr. Evelyn Molloy







The Major Challenge



Believes \rangle Anxiety — Risk — Danger \rangle Change

How To Initiate Change Safely



- 1. Differentiate
- 2. Step-by-Step
 - 3. Mindset
 - 4. Experience
 - 5. Controls

Proper Differentiation

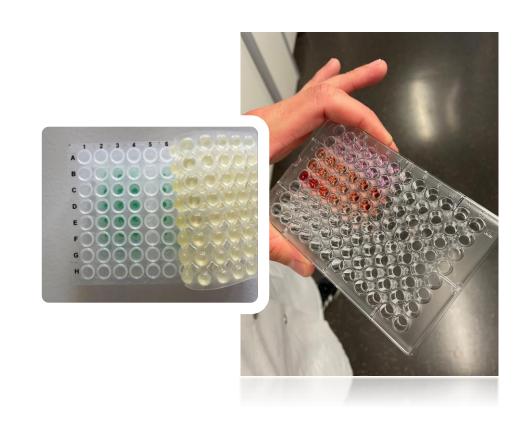


Where | When | How

Samples vs Controls

Quantitative vs Qualitative Data

Preliminary Experiments



Step-by-Step Progression

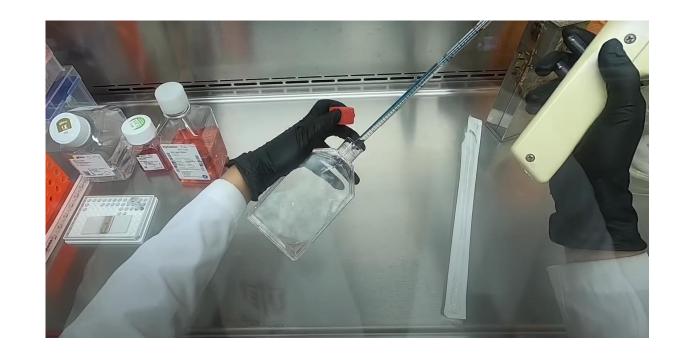


Saving Plastic Waste in S2

Reusing Serological Pipets

Split Multiple Cultures in Parallel

>Avoid Overwhelm + Enable Mistake Spotting



Getting Your Mindset Right

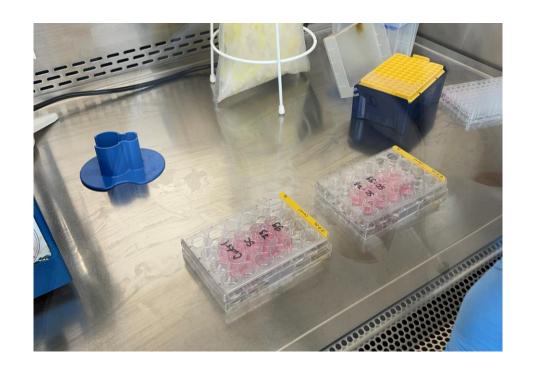


Having A Clear Mind

Be Relaxed

Feeling Motivation

Void of Doubt or Anxiety



Having Sufficient Experience

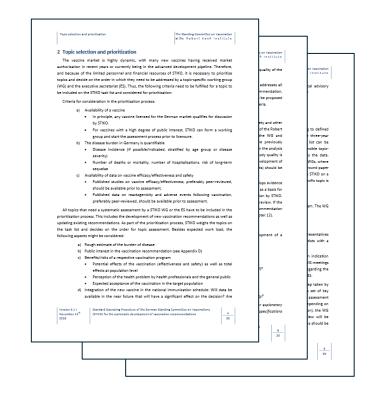


Getting Comfortable

Familiarity with the Protocol

Why versus What

Following Best Practices



Implementing Controls

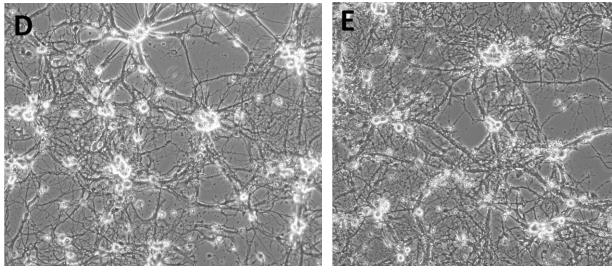


A control for every step

Test all important aspects

Can be general – "Contamination"?

Document Properly



10.1016/j.jneumeth.2007.02.010

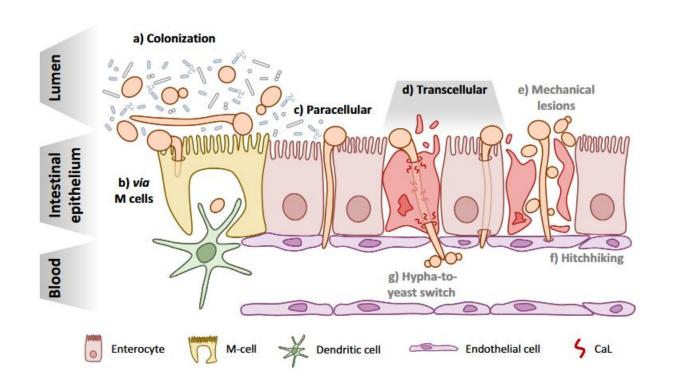


Microbial Pathogenicity Mechanisms - Dr. Jakob Sprague

- Experimental Design
- S2 Environments
- Understanding Limitations



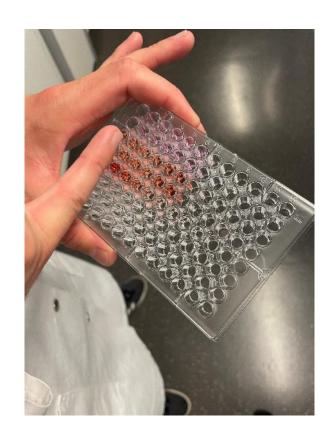
Microbial Pathogenicity Mechanisms - Dr. Jakob Sprague





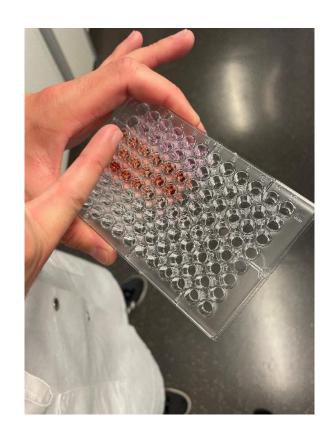
DOI: 10.1080/19490976.2022.2154548









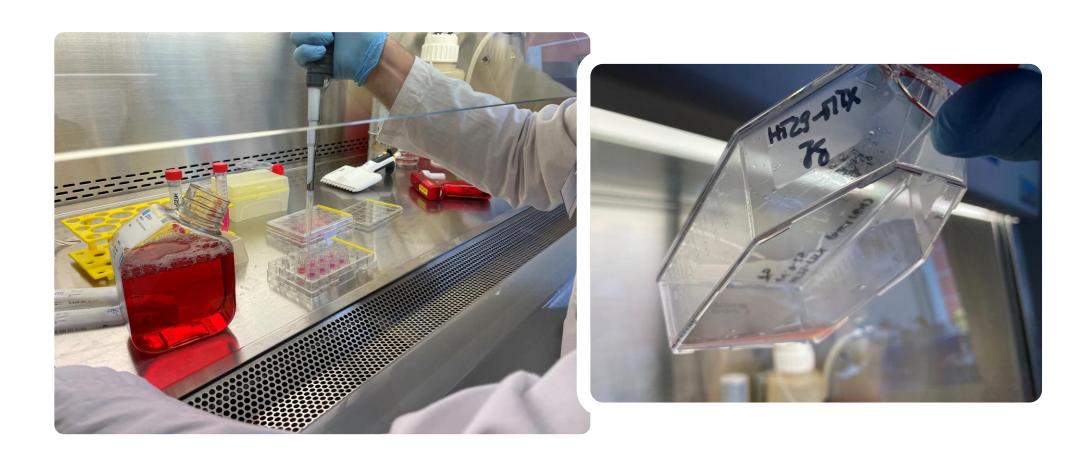






2nd Case Study





You Can Do It Too











Available online at www.sciencedirect.com

Journal of Microbiological Methods 55 (2003) 475-479

Journal of Microbiological Methods

www.elsevier.com/locate/jmicmeth

Note

A 6×6 drop plate method for simultaneous colony counting and MPN enumeration of *Campylobacter jejuni*, *Listeria* monocytogenes, and *Escherichia coli*

Chin-Yi Chen*, Gary W. Nace, Peter L. Irwin

Eastern Regional Research Center, Agricultural Research Service, United States Department of Agriculture¹, 600 E. Mermaid Lane, Wyndmoor, PA 19038, USA

Received 28 January 2003; received in revised form 23 April 2003; accepted 9 June 2003

Journal of Applied Microbiology

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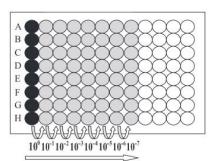
Journal of Applied Microbiology ISSN 1364-5072

ORIGINAL ARTICLE

Nonrecovery of varying proportions of viable bacteria during spread plating governed by the extent of spreader usage and proposal for an alternate spotting-spreading approach to maximize the CFU

P. Thomas, A.C. Sekhar and M.M. Mujawar

Division of Biotechnology, Indian Institute of Horticultural Research, Hessarghatta Lake, Bangalore, India



10-fold serial dilutions (20 µL into 180 µL medium)

Plate six 10 μL-drops from 6 selected dilutions using a multichannel pipette

Incubate at proper temperature

Enumerate colonies

10⁻² 10⁻³ 10⁻⁴ 10⁻⁵ 10⁻⁴ 10⁻⁷

You Can Do It Too



Microbial Immunology - Merle Hammer

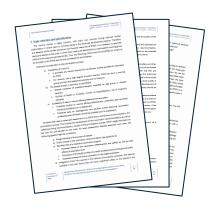




What I Prepared For You



Case Studies



Examples of what your colleagues tested for you already.

Guides

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 Peruing Sylvan tuber notantially after cipring experially for
- frequently used solutions (e.g., Tris solutions when preparing

A checklist-format to inspire and lead you towards change.

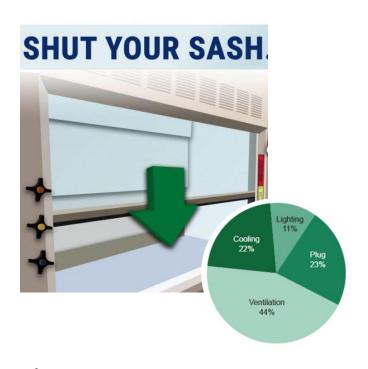
Support



Answering your questions directly and providing you follow-up resources

Saving Potential





Havard | US Davis & Santa Barbara \$4-6000

Saving \$513 Million per University

Getting More Money













Thank You & All Resources





Thank You

Patrick Penndorf Email

LinkedIn

