



THE CENTER FOR
SCIENCE & SOCIETY
AT COLUMBIA UNIVERSITY

GENSPACE



THE FUTURE OF TEXTILE INDUSTRY: BACTERIAL COLORANTS IN FASHION

Dec 14th, 10:30-2:00 pm
Dec 15th, 12:00-4:00pm

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@MinuteBodies

Support for this workshop is made possible by the Public Outreach Grant from Columbia University's Center for Science and Society

WORKSHOP PLAN

DAY 1-Dec 14th, 10:30-2 pm

Cochineal

Talk (30min), Naomi

Hands-on work (1 hour)

- Browse samples
- Crush cochineal, observe under the microscope
- Play with pH (lemon, potash, alum)

Bacteria

Talk (30min), Sümeyye

Hands-on work (1 hour)

- Painting with bacteria

DAY 2-Dec 15th, 12-4pm

Cochineal

Hands-on work (2 hours)

- Textile dyeing

Bacteria

Hands-on work (1 hour)

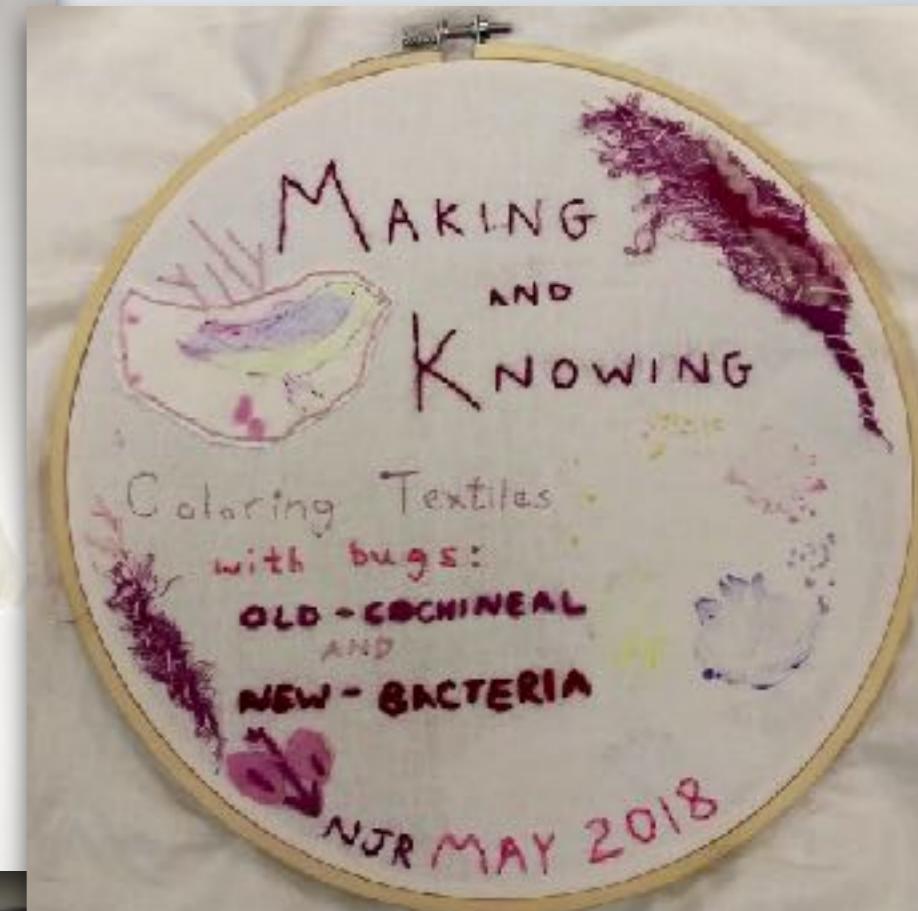
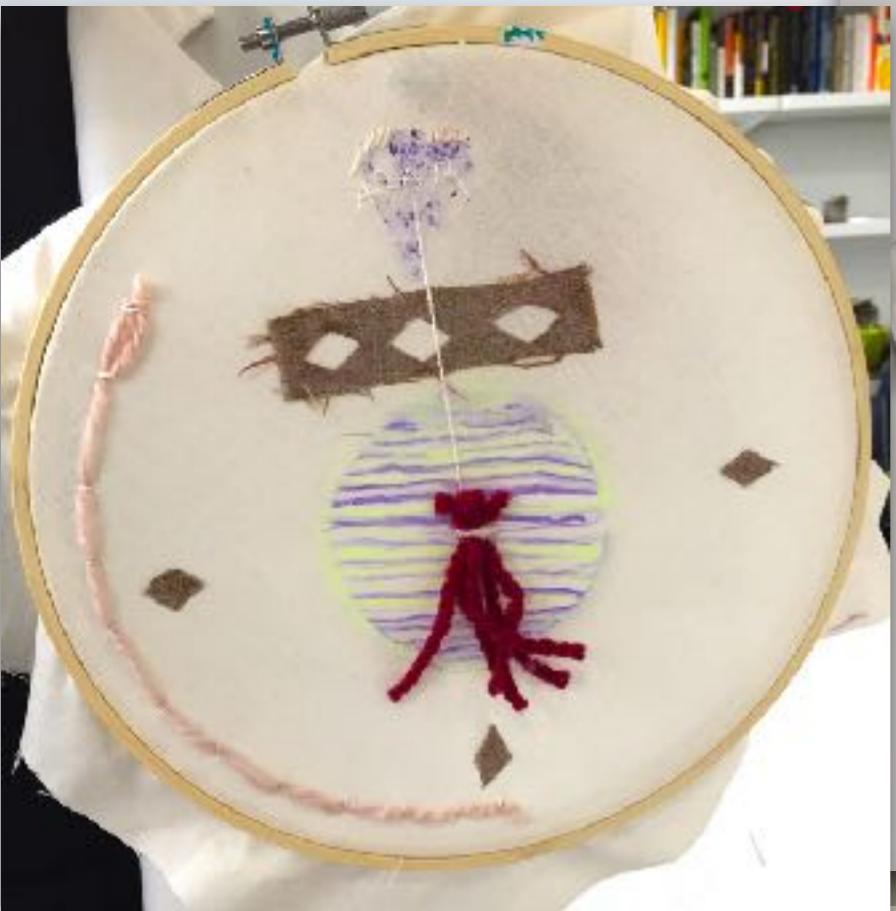
- Printing bacterial designs on textile

Making Bacteria + **Cochineal** Collages

Hands-on creativity time (1 hour)

- Create your own collages by combining bacterial prints with pieces of Cochineal dyed textiles for a final art piece.

SAMPLES FROM PREVIOUS WORKS





Credit: Courtesy of Vlisco

► Natural vs Synthetic Colorants

NATURAL COLORANTS

SOURCES

Plants (e.g. indigo, saffron)

Minerals (e.g. ferrous sulphate, ochre)

Invertebrates (e.g. cochineal, lac insects)



LIMITATIONS

- Not economical for large scale manufacturing
- Colors may tend to fade quickly
- Labor intensive harvesting and dyeing processes
- Requires land for growing plants/insects
- The growth of plants/insects are easily affected by weather conditions, disease, and pests
- Limited supply cannot keep up with “fast fashion”
- Dyeing with some plant-derived dyes requires large amount of water and chemical fixing agents (chemicals!)

ADVANTAGES

- Minimal environmental impact
- Biodegradable
- Obtained from renewable sources
- Most of them are well-known antioxidants
- Healthy choice (but the allergy might be an issue)

SYNTHETIC COLORANTS

- Man-made, chemically synthesized. Discovered in the mid-19th century.

ADVANTAGES

- Colorfast, lightfast and reproducible color
- Limitless color palette
- Easy and fast dyeing process
- Low cost



SYNTHETIC COLORANTS

LIMITATIONS

- Detimental to the environment.
- Many dyes and their breakdown products are carcinogenic, mutagenic and/or toxic to life.
- Due to the stability of dyes, conventional waste treatment methods are inefficient.



Photo: Business Insider / Stringer / Reuters



Synthetic fabric dyes & treatment chemicals

90% of the local groundwater is polluted in China and 72 toxic chemicals in the water supply are from synthetic textile dyeing.



**CURRENT TEXTILE INDUSTRY CAUSES POLLUTION!
CAN MICROORGANISMS HELP?**

USE OF MICROORGANISMS IN BIODEGRADATION OF DYES

Bacteria, yeast, fungi, and algae are already used for the decolorization and degradation of synthetic dye waste. But the process is still far from complete/successful.

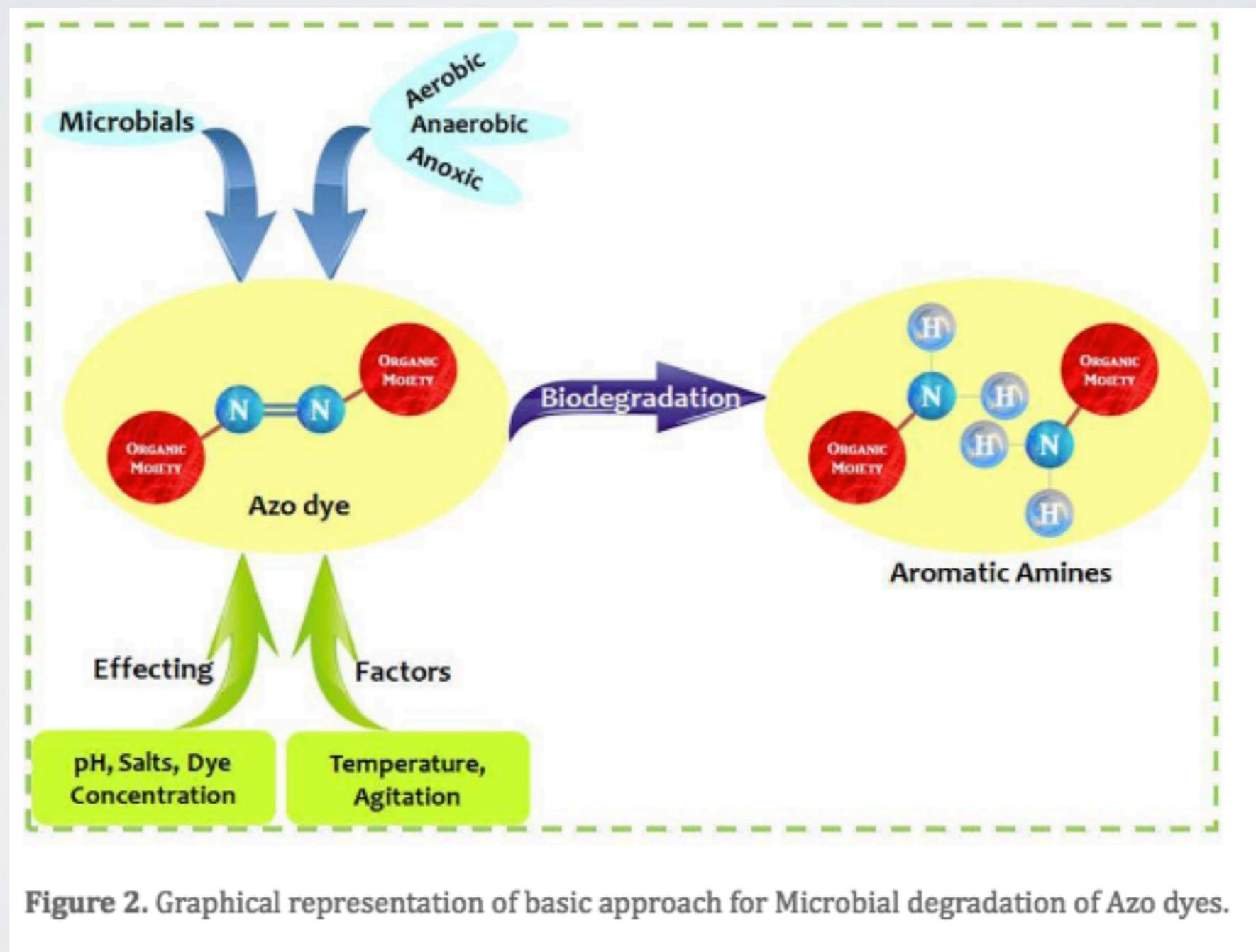


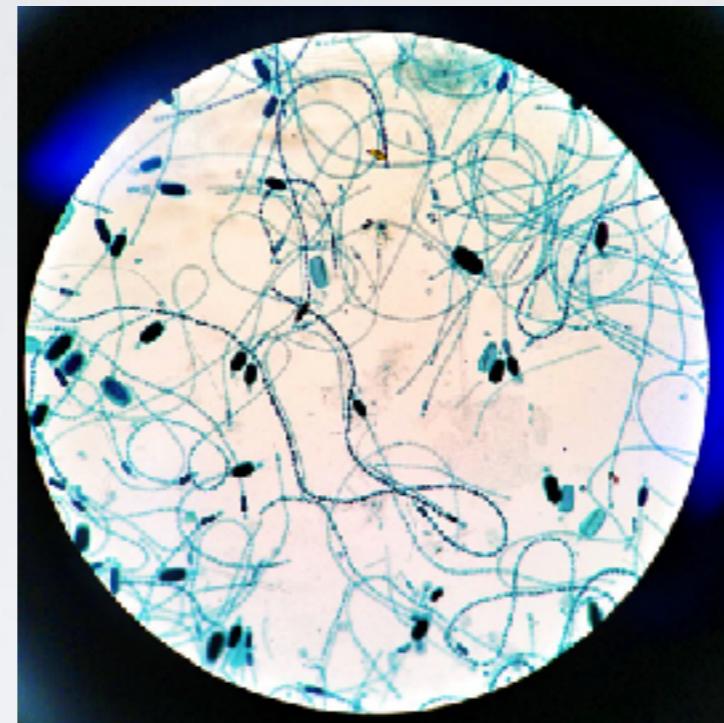
Figure 2. Graphical representation of basic approach for Microbial degradation of Azo dyes.

USE OF NATURALLY COLOURED OR ENGINEERED MICROORGANISMS IN THE PRODUCTION OF SUSTAINABLE AND GREEN COLORANTS

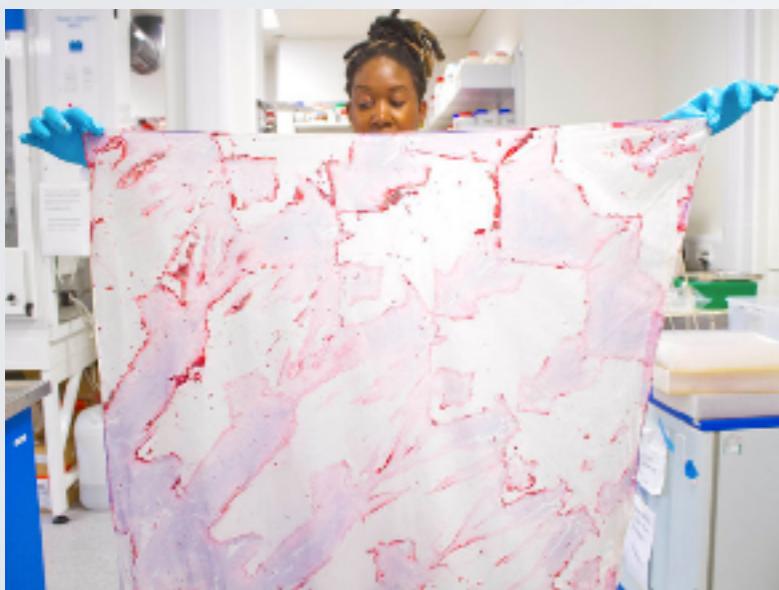
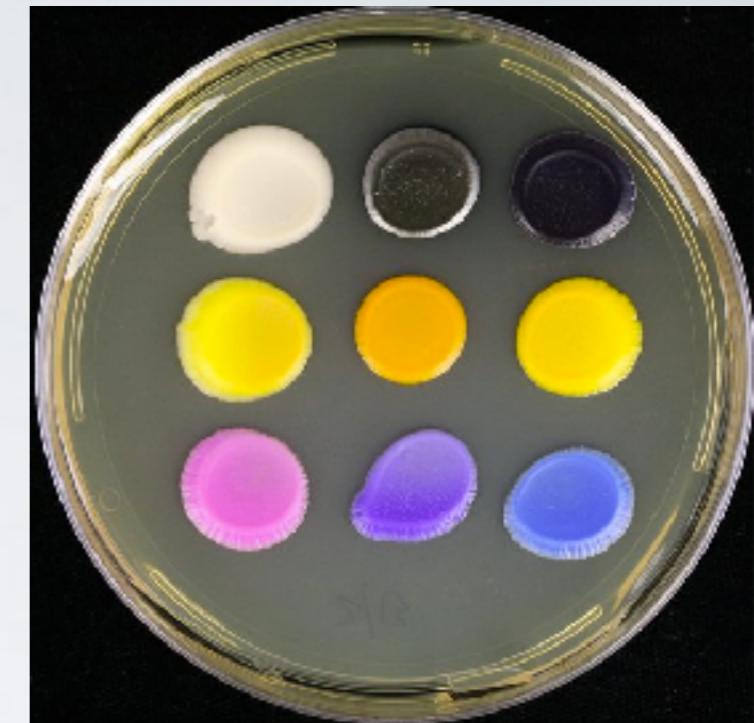
Bacteria



MicroAlgae



Yeast



Natsai Audrey Chieza with one of the fabrics she dyed by using pigment-producing bacteria



Blond & Bieber studio,
color changing algae prints



Jasmine Temple, painting
with yeast

WHY MICROORGANISMS PRODUCE PIGMENTS?

FUNCTION:

1. Photosynthesis (e.g.chlorophylls)
2. UV protection (e.g.carotenoids)
3. Secondary metabolites for storage of energy
4. Stress survival
5. Communication
6. Virulence (e.g. Pyocyanin blue toxin)
7. Antibiotic
8. Antiviral

*Pigments can be water soluble or insoluble

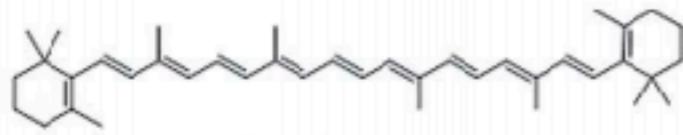
*Some pigments are released from the organism, while some are kept inside the organism

*Oxygen is essential for pigment production thus only aerobic bacteria can produce pigment.

*Pigment synthesis depends on pH, temperature, growth medium constituents, oxygen availability.

Each pigment is different. Hence every coloration process requires diligent optimization!

BACTERIAL PIGMENTS



Carotenoids
(Orange)

Flavobacterium
Sphingobacterium
Actinobacteria
Agrobacterium aurantiacum

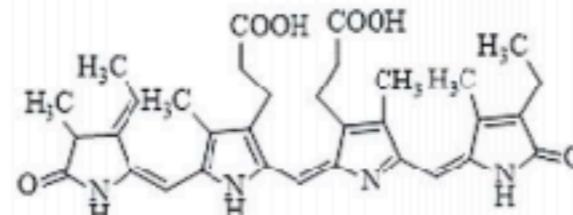
Antioxidants
Food supplement
for humans and as
food additives for
animals and fish.



Flexirubin
(Yellowish-Orange)

Chryseobacterium
Flavobacterium

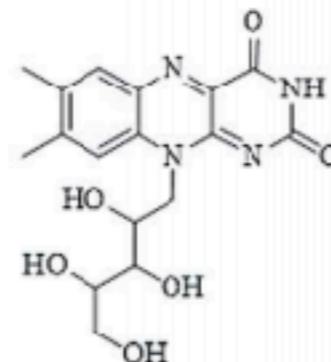
Treatment for
chronic skin
disease, eczema,
gastric ulcers etc.



Phycocyanin
(Blue)

Cyanobacteria

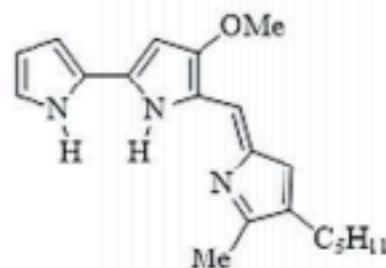
Dietary supplement
rich in proteins



Riboflavin
(Yellow)

Bacillus subtilis

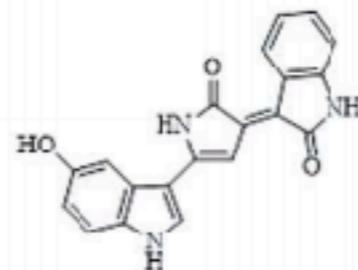
Used in foods,
vitamin enriched
milk products and
energy drinks.



Prodigiosin
(Red)

Serratia marcescens
Pseudomonas
Pseudoalteromonas
Alteromonas denitrificans
Hahella
Vibrio

Antibacterial
Antifungal
Antimalarial
Immunosuppressive
Anticancer



Violacein
(Purple-blue)

Chromobacterium violaceum
Alteromonas luteoviolacea
Janthinobacterium

Antiviral
Antibacterial
Antiulcerogenic
Antileishmanial
Anticancer

Fig. 2 Structure of bacterial pigments.

ADVANTAGES OF USING MICROORGANISMS AS A NATURAL COLORANT FACTORY



An example of
bacterially dyed clothing
by Natsai Audrey Chieza

- Microorganisms are easy to grow and harvest at industrial scale in bioreactors
- They produce a wide array of pigments
- Genetic engineering makes it possible to create limitless color palettes and grow microorganisms faster
- Environmentally friendly
- Biodegradable
- No need to worry about weather conditions or soil quality
- Dyeing processes doesn't usually require any toxic chemicals
- Most pigments have natural antibiotic, anti-cancer, anti-mycotic properties



An example of bacterially printed/
stamped design on the textile



Pigmented microorganisms
growing on the surface of a
agar Petri dish.

CURRENT LIMITATIONS/CHALLENGES IN USING MICROORGANISMS AS A NATURAL COLORANT FACTORY

- Fermentation technology is already in use (brewery tanks) to grow bacteria at the industrial scale; however, more research is needed to increase pigment production. Novel scientific developments in genetic engineering are promising!
- Growing bacteria at an industrial scale is still not cost effective. More research is needed to formulate sustainable and cost-efficient growth medium. A “circular economy” model is ideal.
- Current pigment separation and purification methods are tedious and time-consuming. Some steps require the use of toxic chemical solvents. More research is needed on alternative separation techniques.
- Some pigments are sensitive to light, heat, and pH, and they are insoluble in water. Dyeing conditions will need to be optimised for each different pigment. Some pigments may require stability enhancement through genetic engineering.
- More research is needed to develop methods and tools for ink, print, and pattern making.

EXAMPLES OF BIOTECHNOLOGY COMPANIES PRODUCING NATURAL COLORANTS FROM MICROORGANISMS

Faber Futures

<https://www.fabefutures.com/> ▾

Faber Futures is a biodesign lab fusing design thinking with living biological systems to generate scalable models for sustainable futures.



Pili-Bio

<https://www.pili.bio/> ▾

Through biotechnology, PILI conjugates the performance of the chemical industry with the renewability of biology to cope with the challenges of a clean color ...

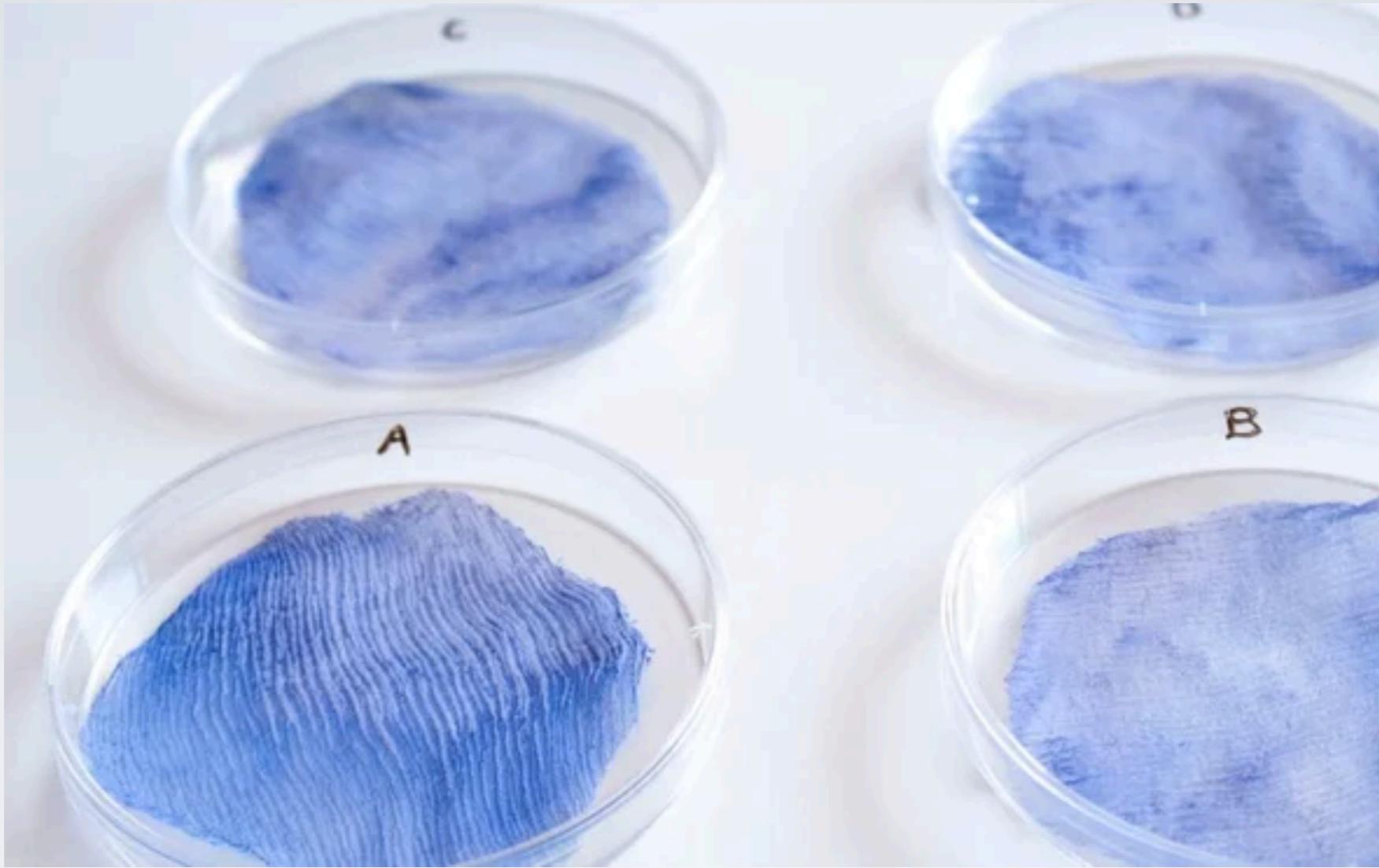
Colorifix | Home

www.colorifix.com/ ▾

Welcome to Colorifix. Colorifix is developing a revolutionary dyeing process to help the textile industry dramatically reduce its environmental impact in a cost ...



And many university laboratories...



Dyeing with Bacteria: Research and Development

Direct dyeing



Co-culturing textile with Bacteria



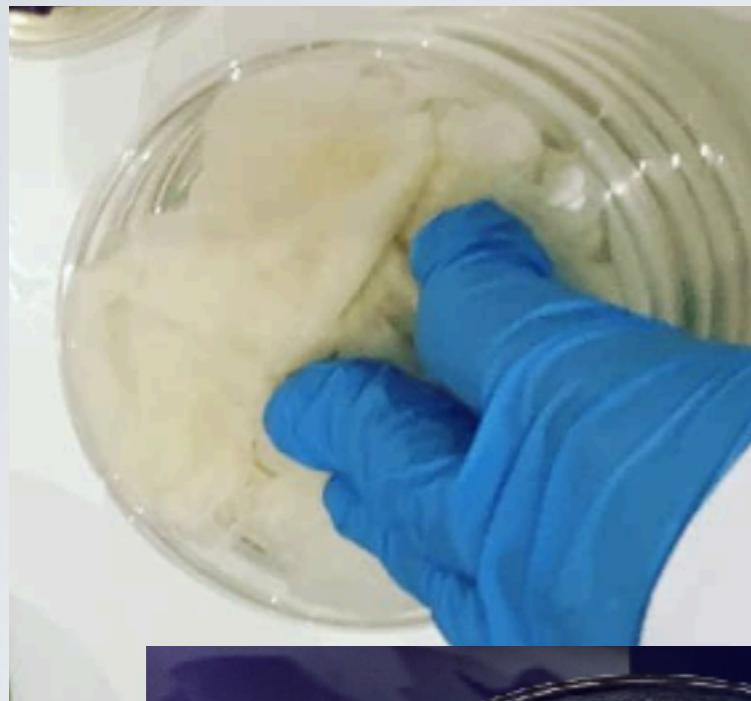
Natsai Audrey Chieza used *Streptomyces coelicolor* bacterial strain for this application.

- 1) Grow Bacteria in liquid or solid medium until it expresses the pigment
- 2) Insert textile into the liquid/solid medium and co-culture until the desired color saturation is achieved
- 3) Autoclave or boil the textile-bacteria mixture to kill bacteria and extract the pigment.
- 4) Wash the textile to remove impurities

This method will only work for bacterial pigments that can directly bind to textile fiber

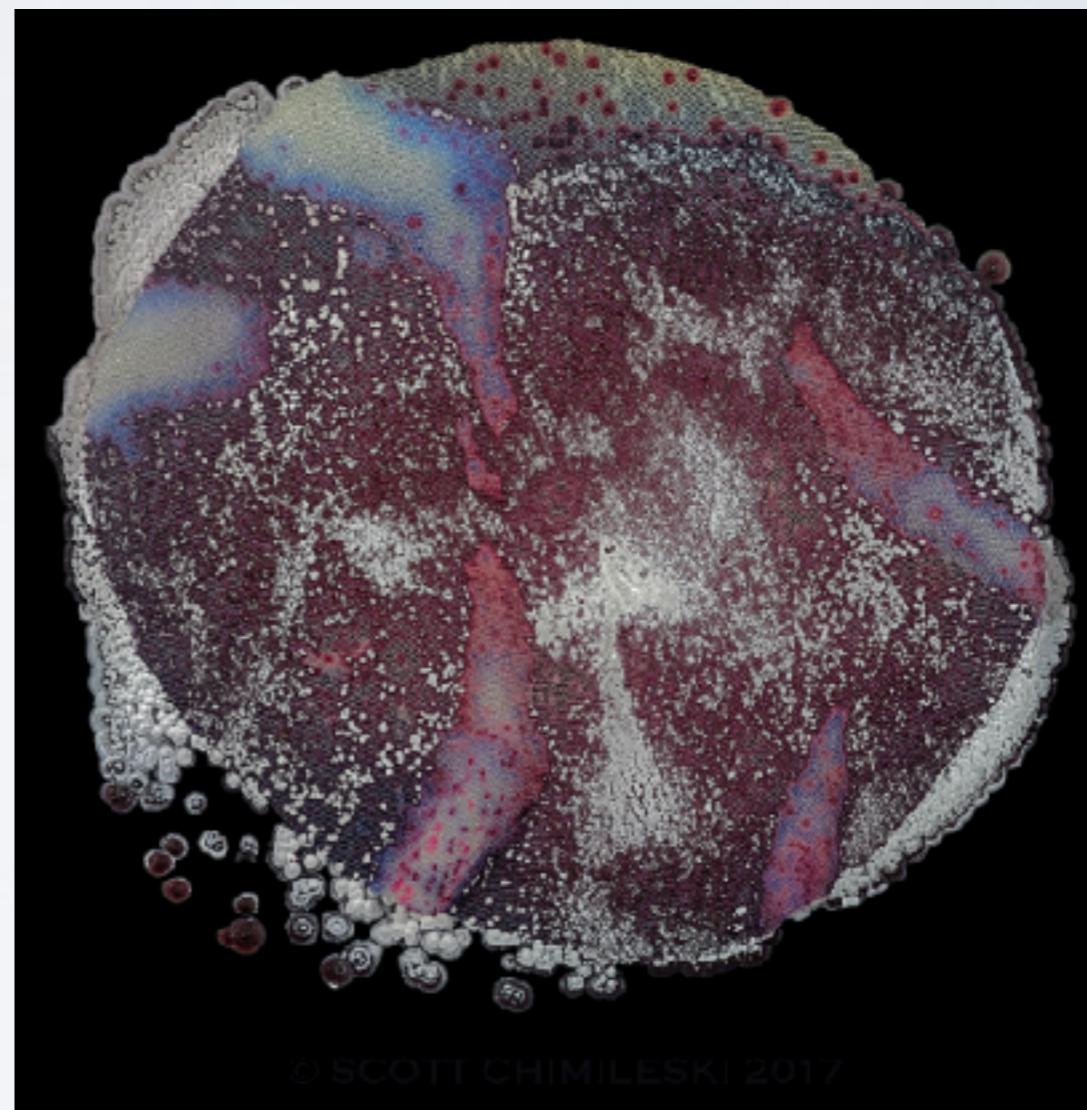
DIRECT DYEING EXAMPLES

(not with isolated pigment but with living bacteria)



Violacein pigment *Janthinobacterium*

- Insert a piece of fabric into a Petri dish with already grown bacteria in it or pour some liquid bacterial culture over the textile, wait for bacteria to grow and do the magic!



© SCOTT CHIMILESKI 2017

A small piece of fabric fit for a Petri dish and dyed during incubation with *Streptomyces coelicolor*.

DIRECT DYEING EXAMPLES

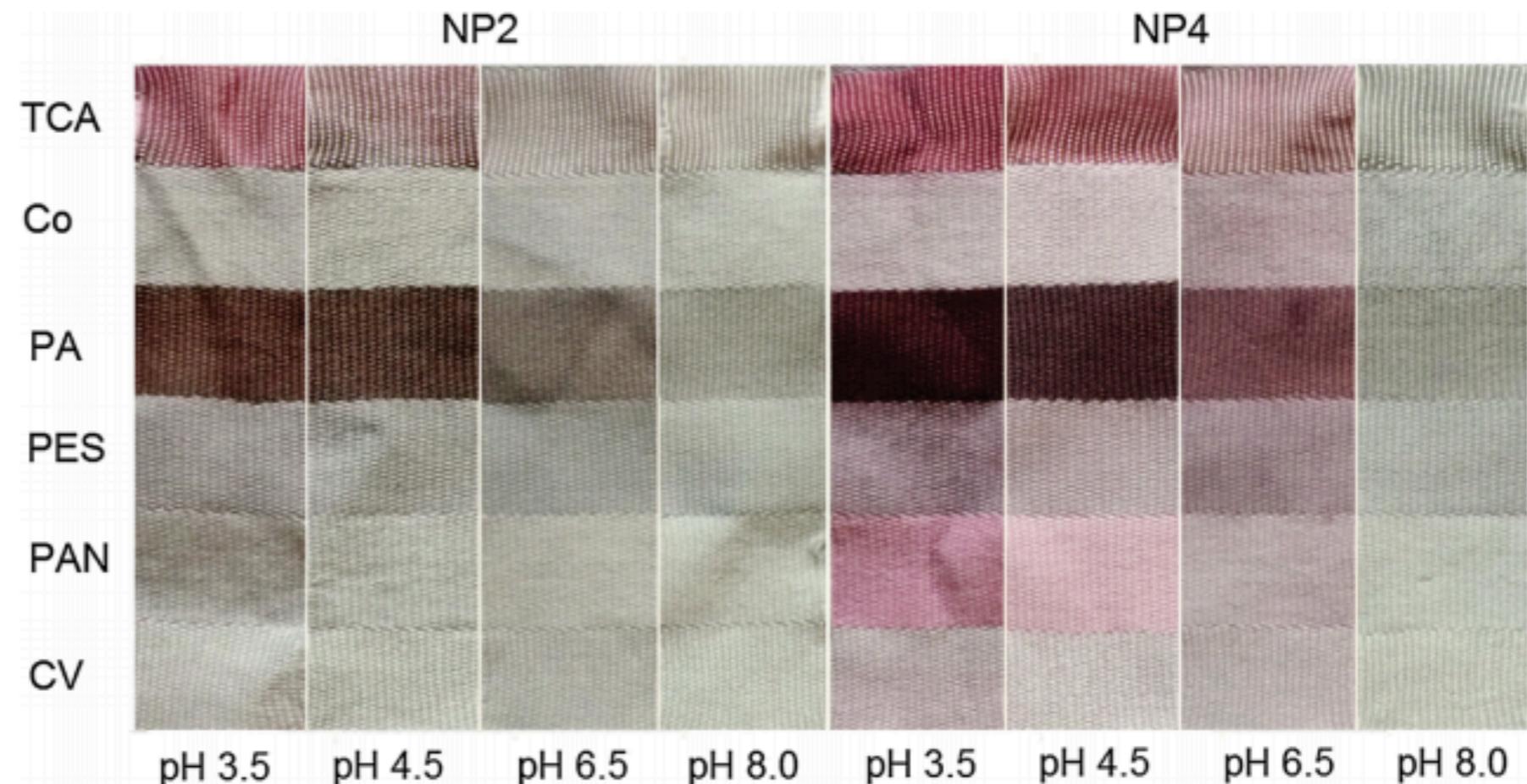
Harmless soil bacteria of the *Streptomyces*

World J Microbiol Biotechnol (2014) 30:2231–2240
DOI 10.1007/s11274-014-1644-x

ORIGINAL PAPER

Crude bacterial extracts of two new *Streptomyces* sp. isolates

Fig. 6 Multifiber fabric dyed with pigment NP2 and NP4 in the dyebaths of different pH (3.5; 4.5; 6.5 and 8.0). *TCA* triacetate, *Co* bleached cotton, *PA* polyamide, *PES* polyester, *PAN* acrylic, *CV* viscose



Mordant dyeing



Co-culturing
mordanted textile with
Bacteria

- 1) Grow Bacteria in liquid/solid medium until it expresses the pigment
- 2) Insert mordanted textile into the liquid/solid medium and co-culture until the desired color saturation is achieved
- 3) Autoclave or boil the textile-bacteria mixture to kill bacteria and extract the pigment.
- 4) Wash the textile to remove impurities

**Sometimes the mordant is applied to the textile after dyeing.

**Tamarind and alum are some commonly used mordants in these applications.

AN EXAMPLE FOR MORDANT DYEING WITH BACTERIA

Violacein from *chromobacterium violaceum*

- The fabrics are dyed using the boiling technique with alum used as mordant,
- In the boiling method, 1 g of fabric is immersed in 20 mL of bacterial culture broth and heated at 80 C (1 h)
- Upon cooling, the dyed fabrics are washed with cold water

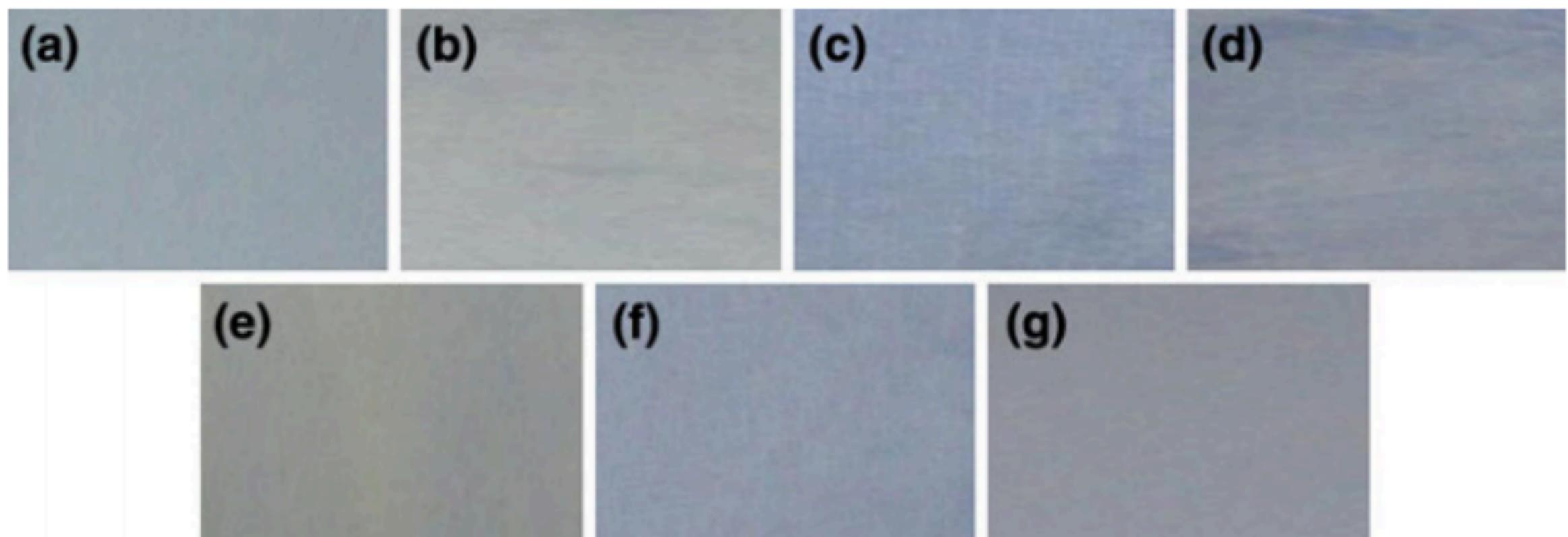


Fig. 4.1 Dyeability of violacein on different fabrics using alum as mordant; (a) pure cotton (PC), (b) pure silk (PS), (c) pure rayon (PR), (d) rayon jacquard (RJ), (e) silk satin (SS), (f) cotton (C) and (g) polyester (P)

DIFFERENT TECHNIQUES FOR PATTERN DESIGN WITH BACTERIA

FABRIC
PREPARATION
METHOD



Printing



Painting



INOCULATION
METHOD

** Each bacterial pigment is different. Different techniques work with different Bacterial pigments.

PART II: INTRODUCTORY INFORMATION FOR THE HANDS-ON PART

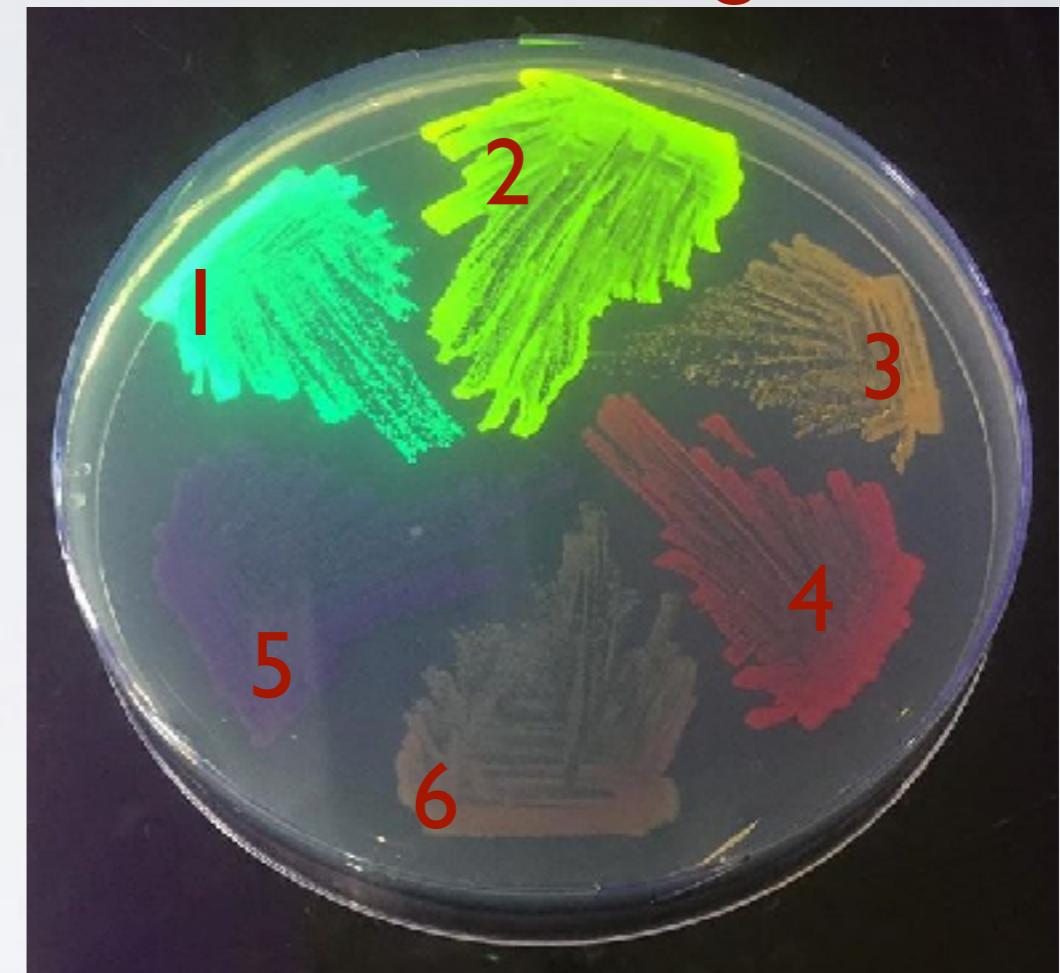
Day I: Painting with bacteria on agar Petri dishes
Day II : Printing/Stamping the designs on textile

WE WILL USE A SAFE E.COLI STRAIN FOR PAINTING/ PATTERN DESIGN

Day-light



Under UV-light

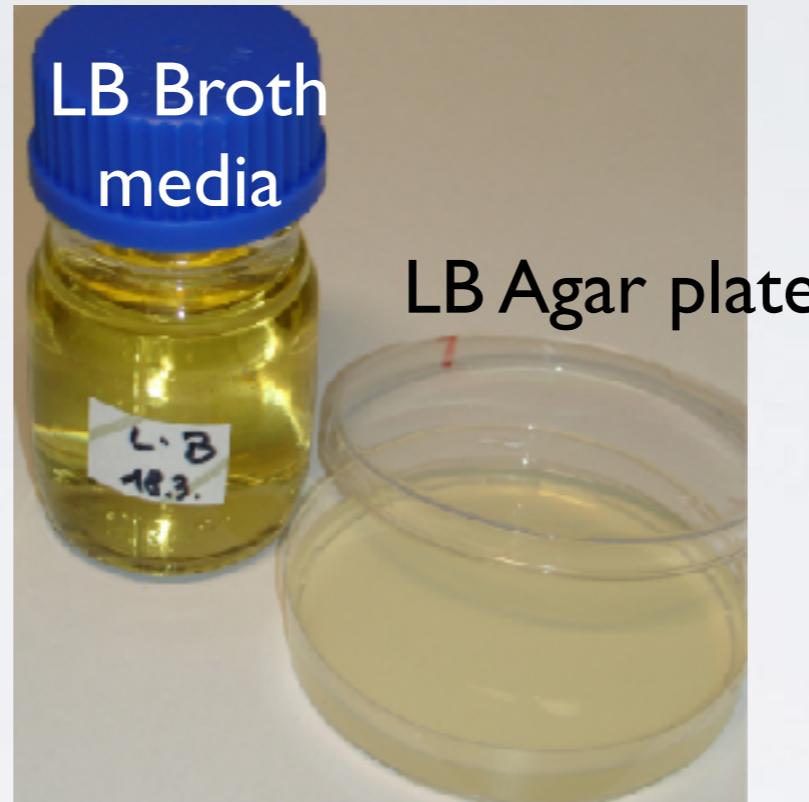


1. Green: very bright chromoprotein (derived from sea anemone)
2. Yellow fluorescent protein: mutated green fluorescent protein (jelly-fish derived)
3. Orange fluorescent protein: mutated red fluorescent protein
4. Red fluorescent protein: (derived from coral)
5. Purple: chromoprotein
6. Blue: chromoprotein (derived from coral)

Bacteria from: petri-paint.com

PREPARATION (already done for you)

I. Prepare Growth Medium: Luria Broth* (LB) Agar Solid Media and LB Broth Liquid Media.



More info here:
<https://www.addgene.org/protocols/pouring-lb-agar-plates/>

LB* is a nutrient-rich media commonly used to feed and grow bacteria in the lab. The addition of **agar** (a gelatinous substance obtained from various kinds of red seaweed) to LB results in the formation of a gel that bacteria can grow on. Bacteria are unable to digest the agar but can gather nutrition from the LB within.

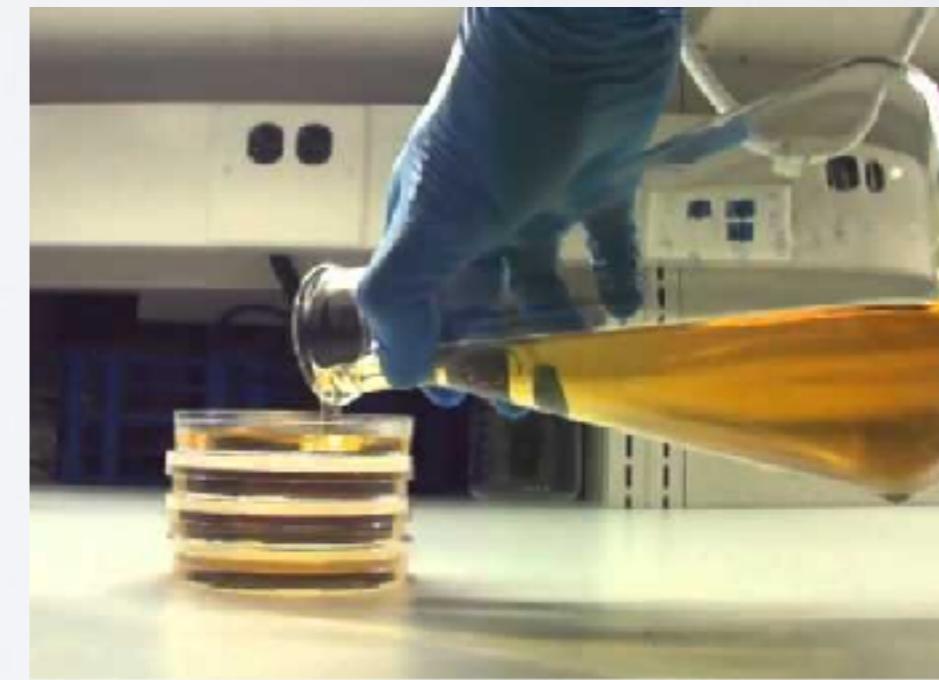
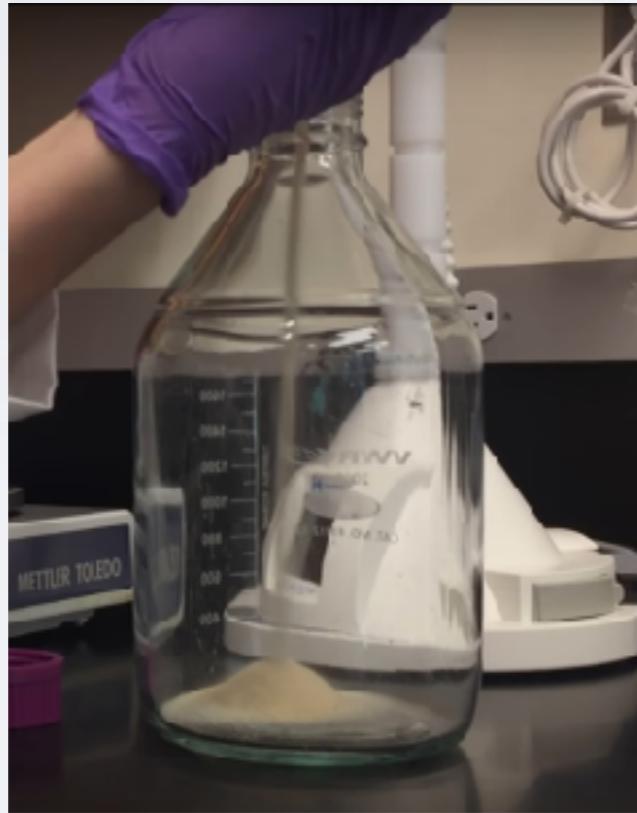
PREPARATION (already done for you)

How to prepare Growth Medium:

- Weight 15 g of LB (luria broth)* agar (for Plates) or 15g of LB (for liquid media) into 1L of water, mix well and autoclave to sterilise.

!LB agar remains liquid above 65 C

- Add antibiotic (35 ug/uL chloramphenicol)
- Pour agar plates, let them cool down and solidify.
- Store at 4C



DAY I: PAINTING/PATTERN DESIGN WITH BACTERIA

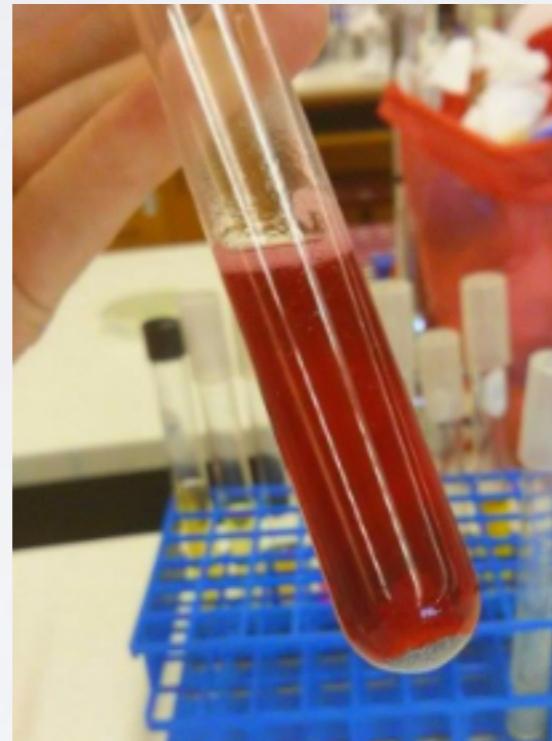
Create your masterpiece!

Paint Palettes

Solid Paint



Liquid Paint



Painting tools

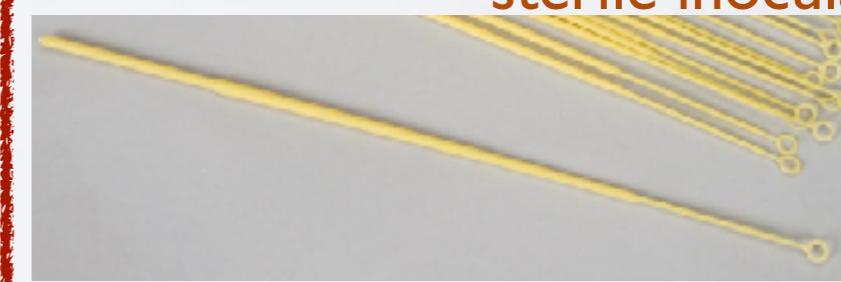
brush



sterile pick



sterile inoculation loop



PAINTING AND PRINTING WITH BACTERIA

Day I. PAINTING-*Create your masterpiece!*

Warnings!

- Wear gloves! Do not touch your face or phone with your gloves!
- Make sure you sterilize your brushes in rubbing alcohol and dry before each use
- If a brush stroke is made in error, you can use as an eraser a paintbrush dipped in rubbing alcohol.
- Use picks gently. Try not to poke the agar
- Use a separate paintbrush for each strain/color to avoid contamination (or dip the brush in alcohol before using for a different color)
- Feel free to experiment. You may try overlaying different colors on top of each other.

Day II. PRINTING/STAMPING



INSPIRATIONAL EXAMPLES OF AGAR ART



microbial communication between the mother and the child within her womb. Some microbes were isolated from the artist, by pressing an agar plate on to her breast. The white Bacillus at the edges of womb were isolated from the hand of the 1-year old daughter of the artist

INSPIRATIONAL EXAMPLES OF AGAR ART



This three-dimensional agar art is a representation of our microbial faces. It was made by pouring agar into a plastic face mask and inverted.

INSPIRATIONAL EXAMPLES OF AGAR ART



The battle of two microbes, as the battle of two seasons. On one side *Staphylococcus*, white as winter snow and *Bacillus mycoides*, they grow fast and cover every other microbes, but when they meet beautiful spring flowers, made by *Serratia marcescens*, they retreat, because antibiotic, produced by *Serratia* inhibit their growth.

INSPIRATIONAL EXAMPLES OF AGAR ART



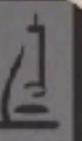
UV radiation was used to duplicate a printed image onto the agar! Principally, UV radiation kills cells by damaging the DNA. If the damage is too widespread, it leads to cell death and hence hinders bacterial growth. Using this theory, we spread bacteria uniformly onto BPA to generate a bacterial lawn. Then, we placed the image that we chose over the agar and exposed it to UV radiation. Only parts that were shaded by the opaque image would allow the bacteria to cultivate, hence forming the image.

INSPIRATIONAL EXAMPLES OF AGAR ART



After bacteria have grown, some were removed/killed by using a sterile brush dipped in rubbing alcohol.

PAINTING TIME!



asm.org/agarart



asm.org/agarart