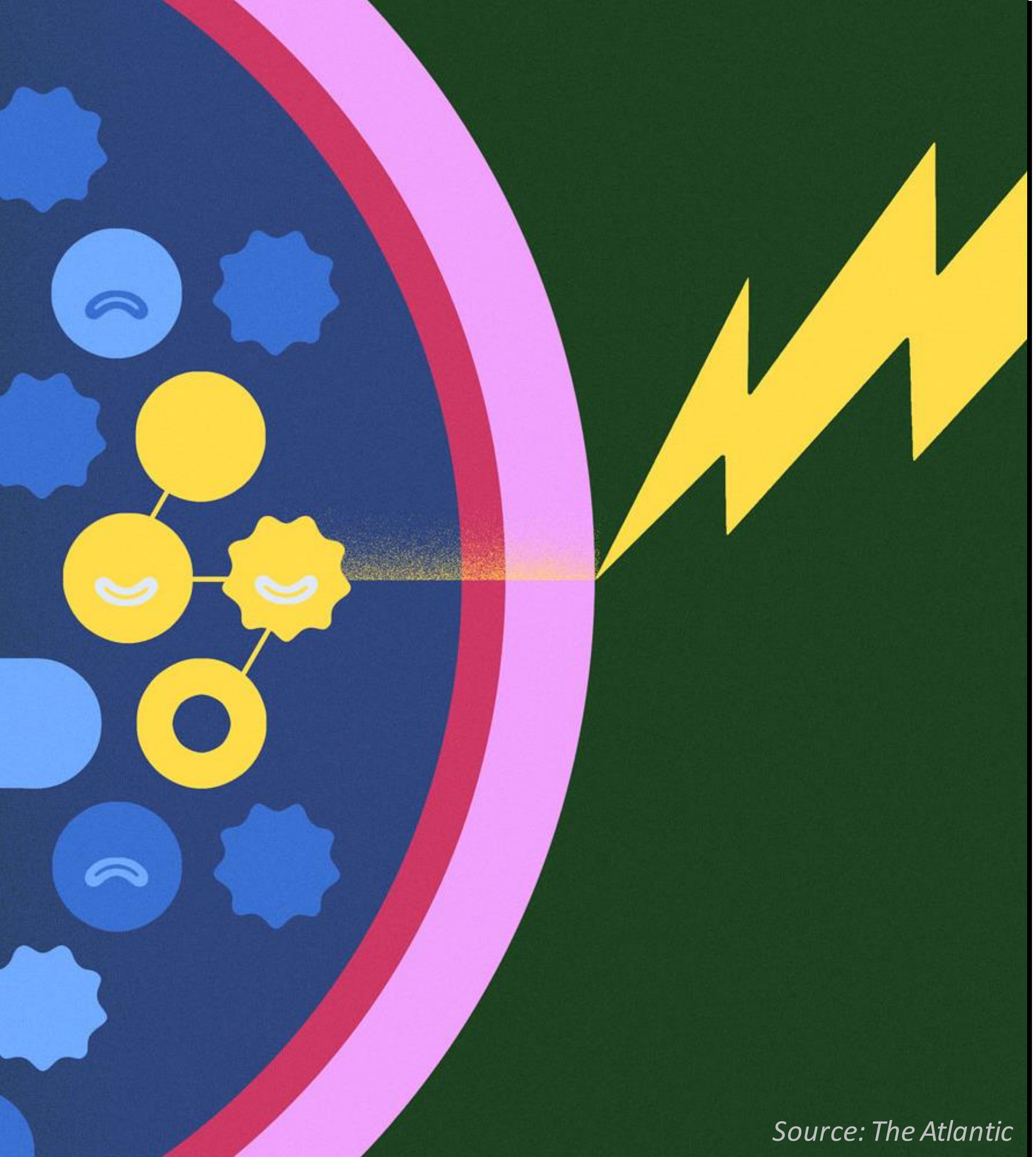
Source: The Atlantic

17.5.2021

Bioelectronic Control of a Microbial Community

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Kim Kutvonen, Maria Heikkilä &
Sami Seppänen*

Aalto University
School of Chemical
Engineering

"What if your fitness monitor or smart watch could detect an active virus in your body – such as COVID-19 – and alert you in real time?

Or, what if a diabetes patient could tap similar technology to administer a dose of insulin as soon as their blood glucose level spikes?"

nature nanotechnology

ARTICLES

https://doi.org/10.1038/s41565-021-00878-4

Check for updates

Bioelectronic control of a microbial community using surface-assembled electrogenetic cells to route signals

Jessica L. Terrell[1,7], Tanya Tschirhart[2,7], Justin P. Jahnke[1], Kristina Stephens[3,4,5], Yi Liu[3,5], Hong Dong[1], Margaret M. Hurley[6], Maria Pozo[3], Ryan McKay[3,4,5], Chen Yu Tsao[3,4,5], Hsuan-Chen Wu[3,5], Gary Vora[2], Gregory F. Payne[3,4,5], Dimitra N. Stratis-Cullum[1] and William E. Bentley[3,4,5] ✉

Terrell et al., Nat. Nanotechnol. (2021)

Outline

1. Introduction
2. Approach
3. Discussion

1. Introduction

Aims & Background

Electronics meets biology

- Biological information transmitted through ions and small molecules while electronic systems require electron flow
- Recently, redox-active molecules were introduced as an option to link electronics with biology
  - Electron flow is coupled to interconversion between biologically distinct redox states

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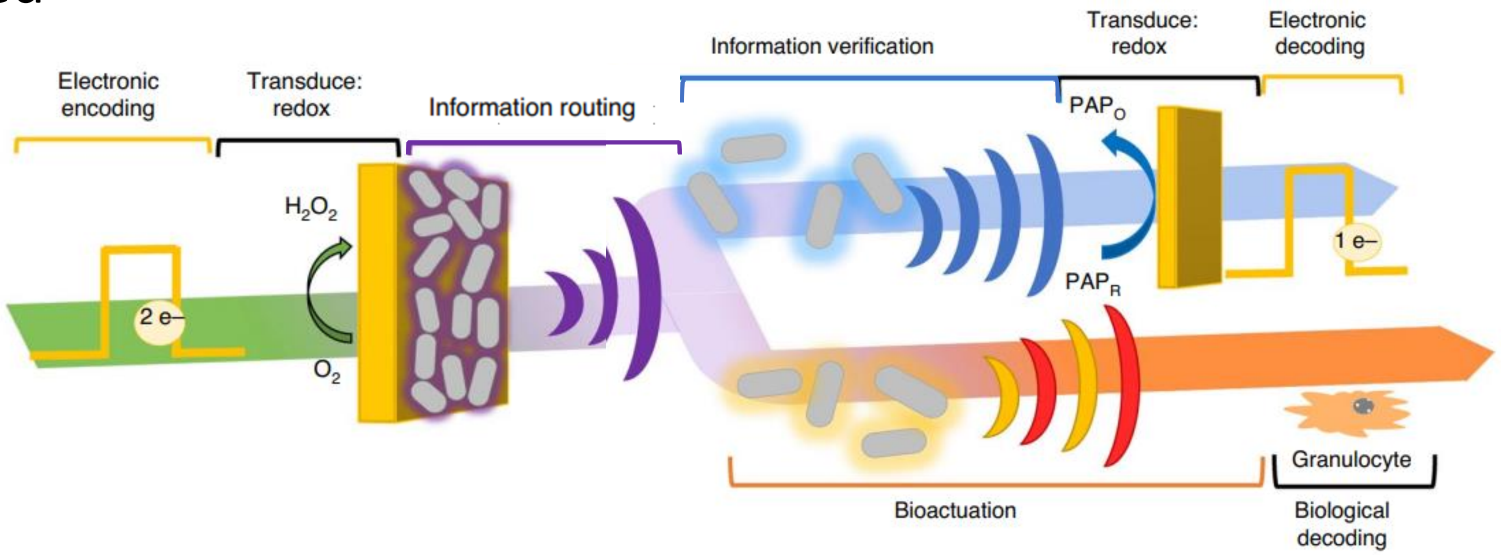
The present study

- Electrode-generated redox signals:
  1. Initiate transduction
  2. Propagate and validate information flow between the electronic system and a community of engineered microbial cells
  3. An electronically controlled biological local area network, "BioLAN", is created



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Main Aims

1. Convert an electronic signal to a biological one via the partial reduction of oxygen
2. Return a verification signal to the electronic interface when the product is expressed
3. Produce target product

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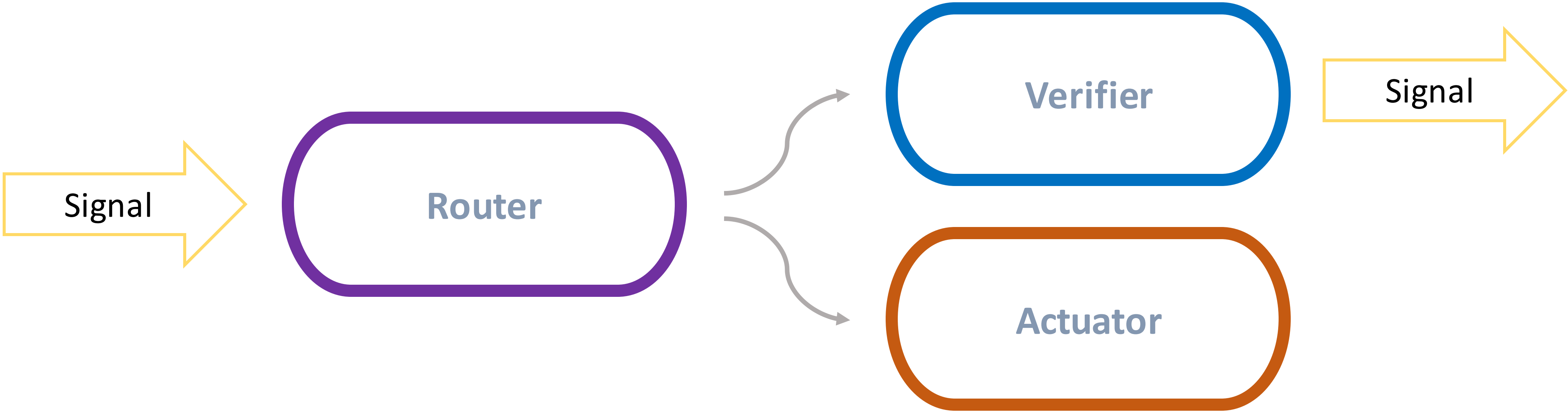
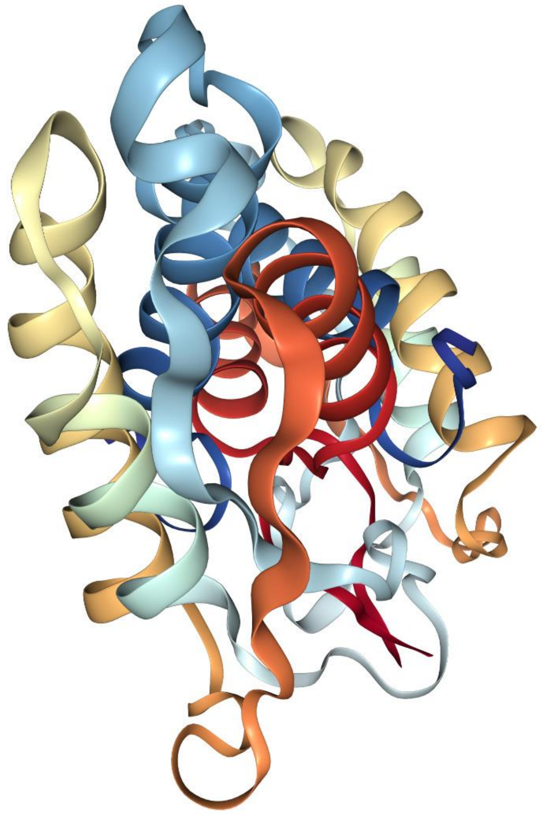
BioLAN Community – Three cell types



Granulocyte Macrophage Colony-stimulating factor (GMCSF)

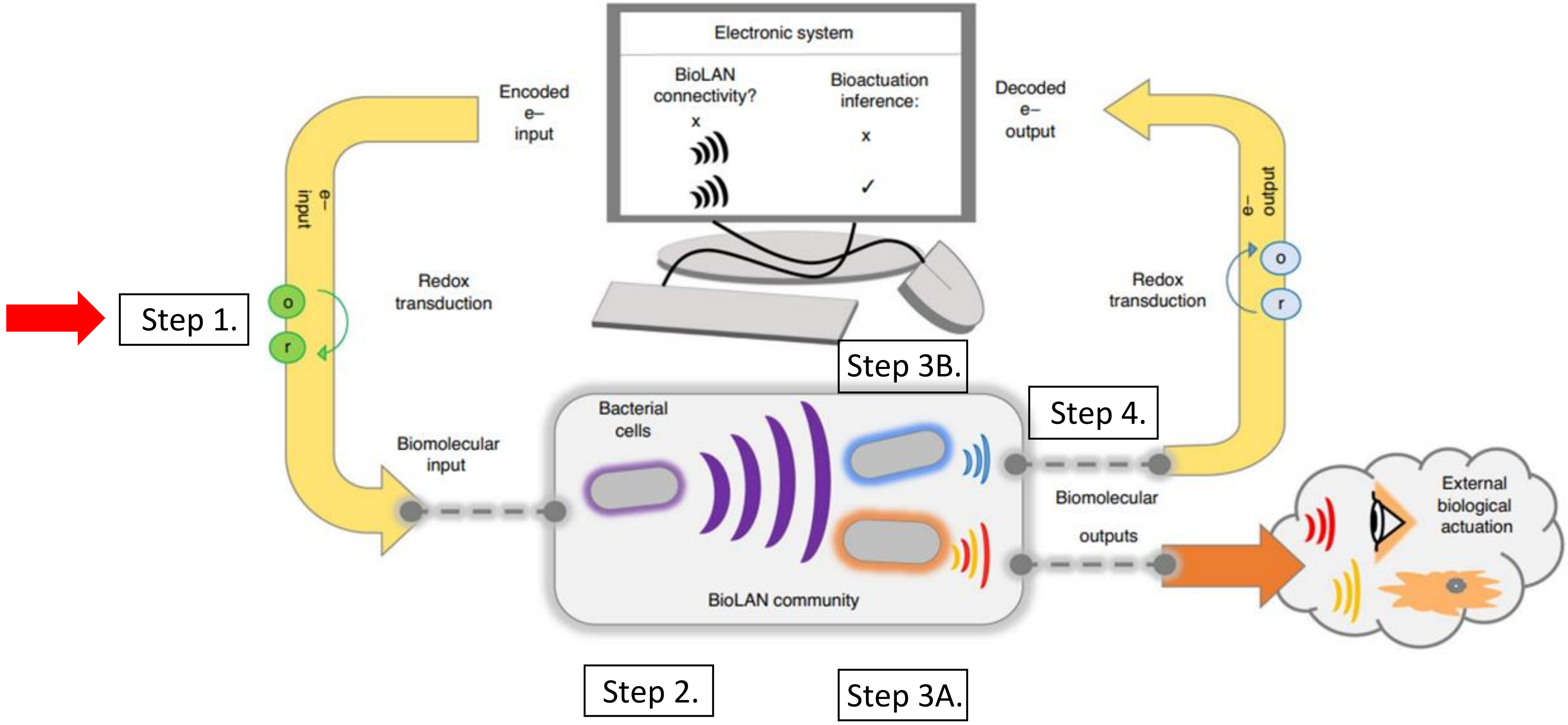
- Cytokine - Major role in immune system
- Pro-inflammatory
- Treatment for Crohn’s disease
- Example product – Can be replaced with e.g., another therapeutic



Source: Sinobiological

2. Approach

4 key steps



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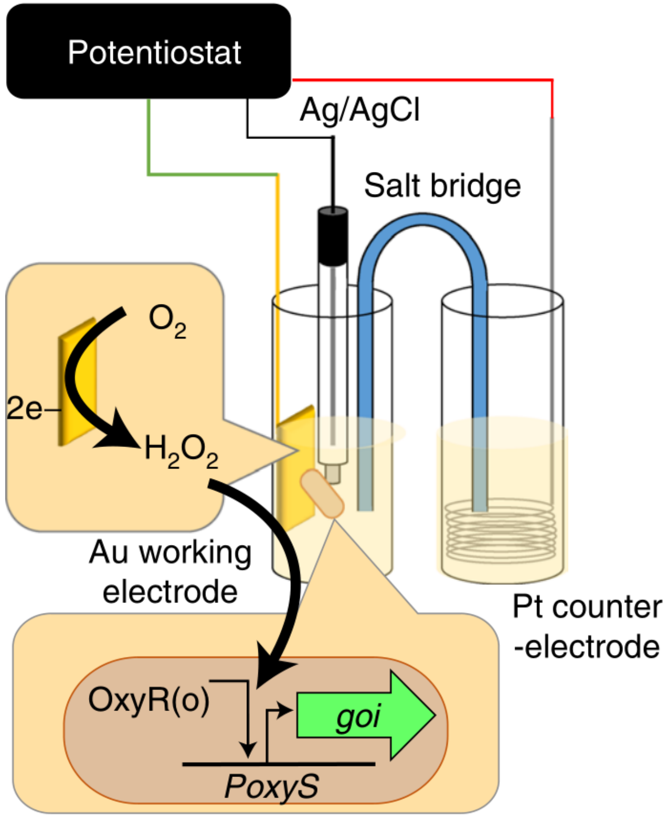
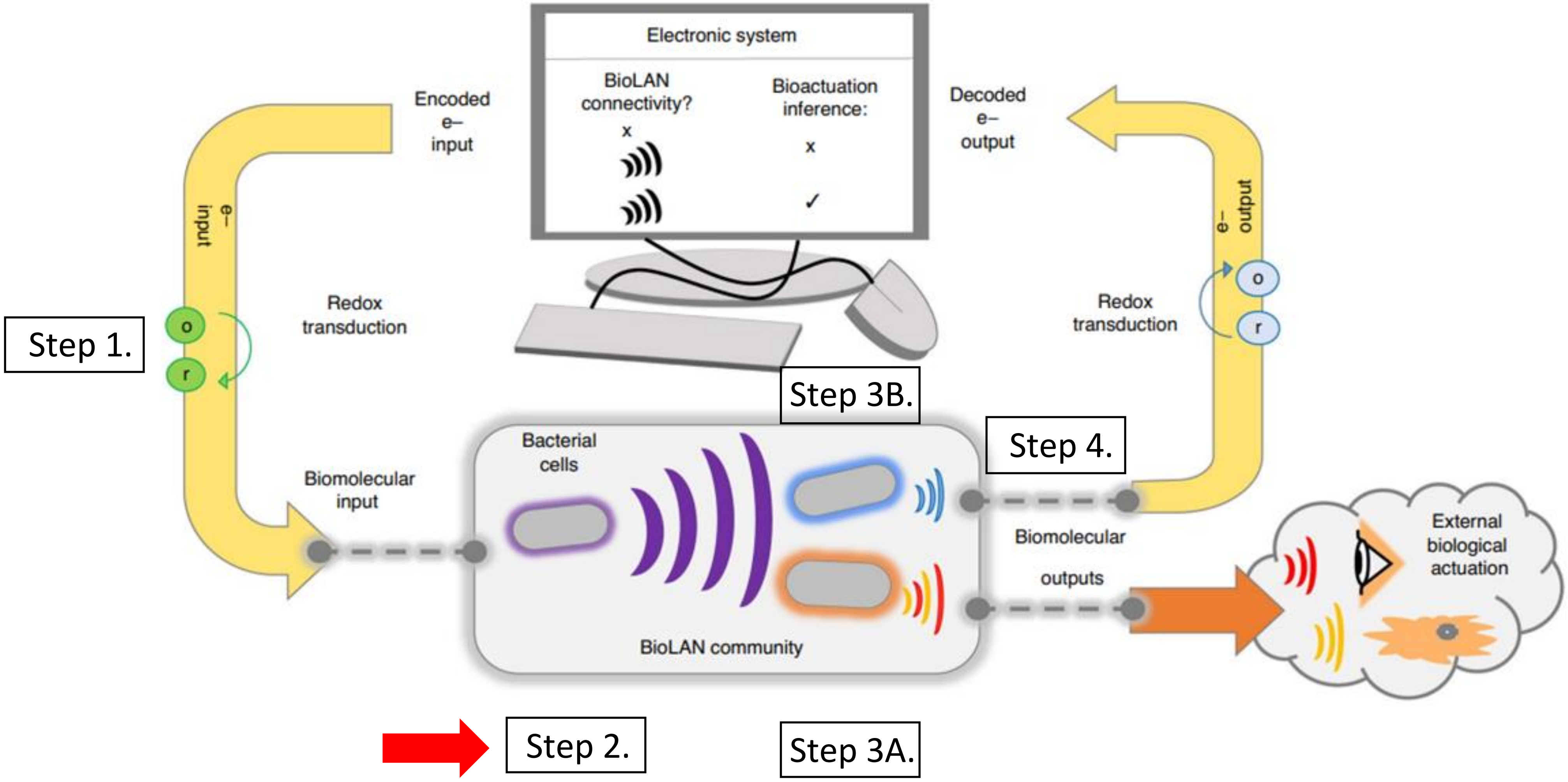
Step 1: Electrochemical reduction of oxygen

- Converts an electronic signal to a biologically recognized one
  - Achieved via the partial reduction of oxygen into hydrogen peroxide
- Router cell is assembled onto gold electrode via high-affinity gold binding peptides



OxyR = Transcriptional activator
PoxyS = Promoter
goi = Gene of interest



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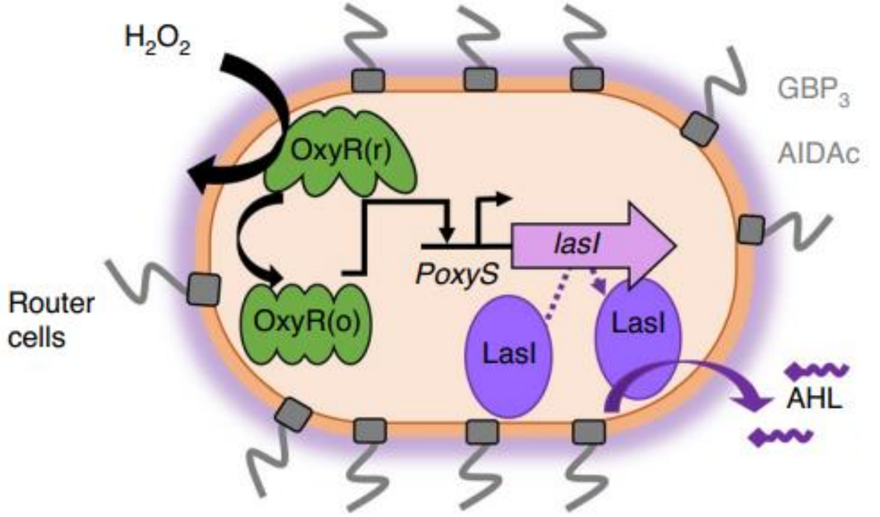
Step 2: Synthesis of signal molecule (AHL)

- Hydrogen peroxide signals the activation of OxyR inside router cell
- OxyR binds to the promoter of *LasI*
- LasI is expressed which then synthesizes AHL
- AHL transfers the signal to two different cell types:
  - Actuator cells
  - Verifier cells



AHL = Signal protein
OxyR = Transcriptional activator
LasI = AHL synthase
AIDAc = Autotransporter pore-forming protein
GBP_3 = Gold-binding peptide
H_2O_2 = Hydrogen peroxide

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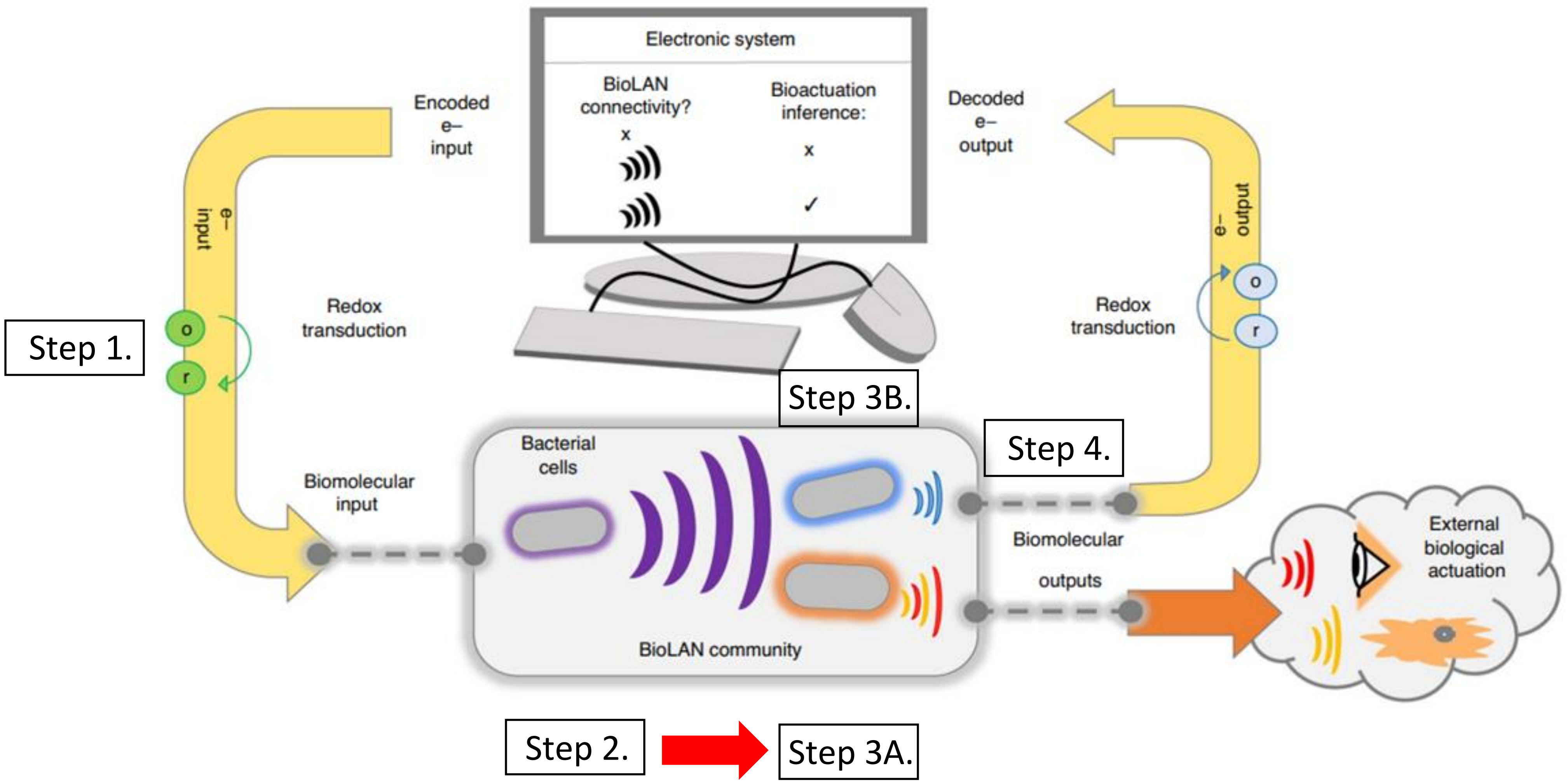

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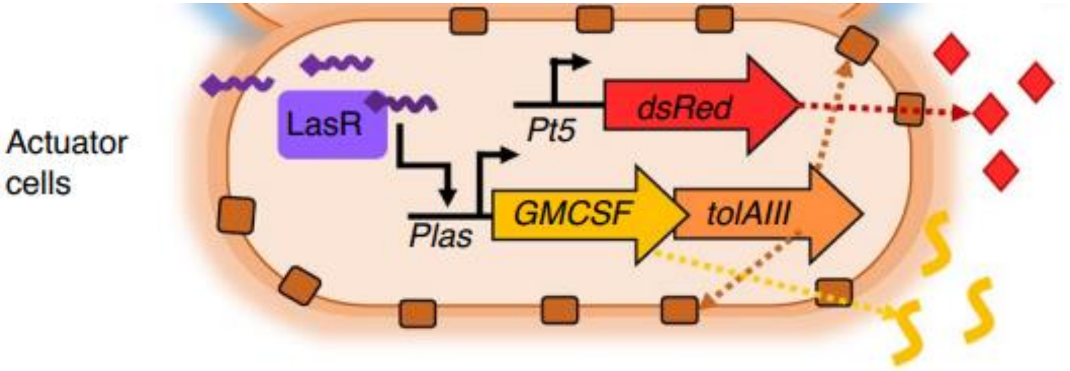
Step 3A: Product synthesis (Bioactuation)

- Bioactuator cells are induced by AHL
  - AHL activates LasR which binds to the Plas promoter
  - Activates the synthesis of GMCSF and *tolAIII*
- *tolAIII* mediates membrane porosity
  - *GMCSF* and *dsRed* are secreted by a cell



LasR = Transcriptional activator
TolAIII = Transmembrane protein gene
Pt5 & Plas = Promoters
GMCSF = Granulocyte Macrophage Colony-stimulating factor
DsRed = Fluorescent protein

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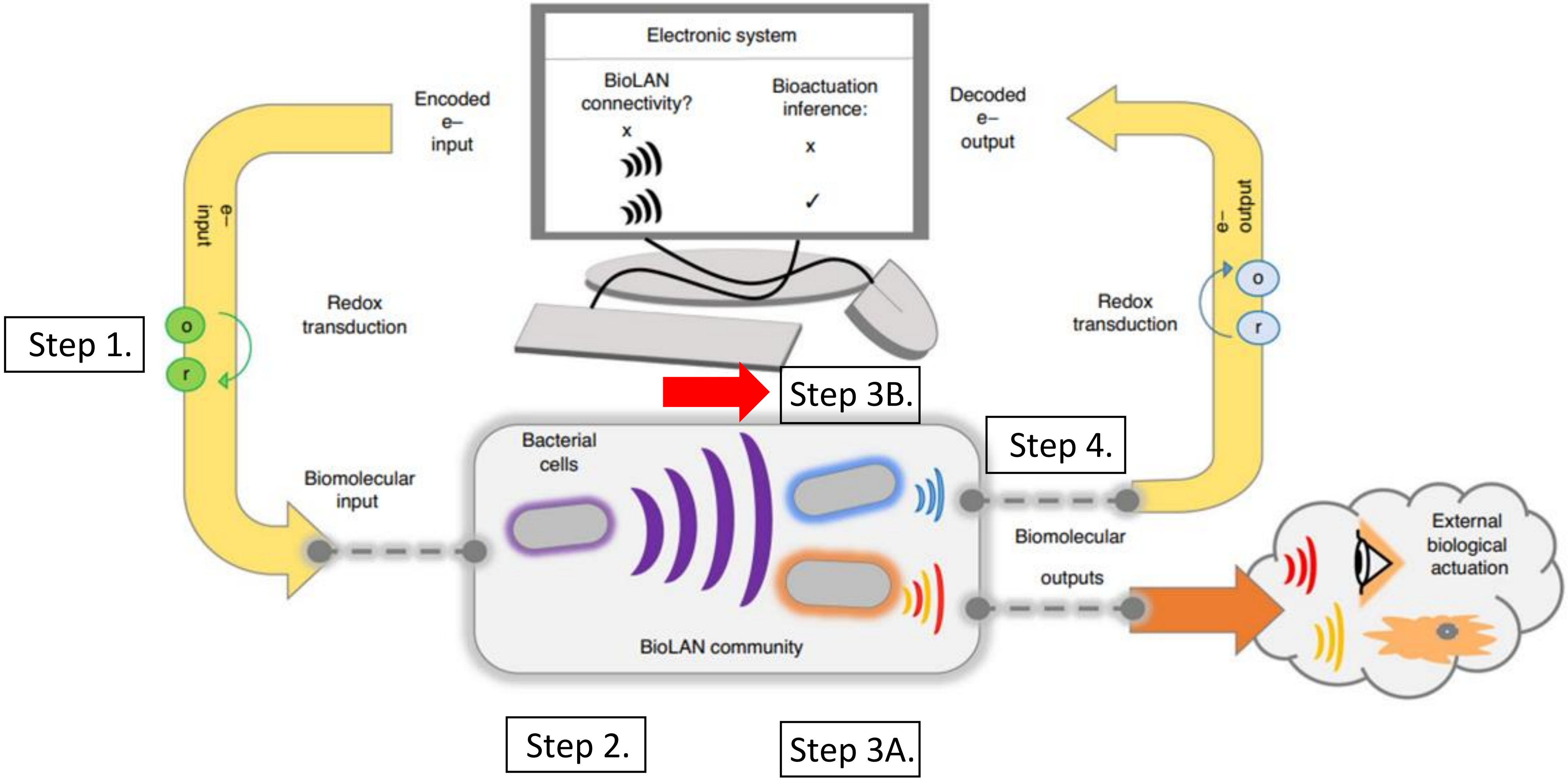

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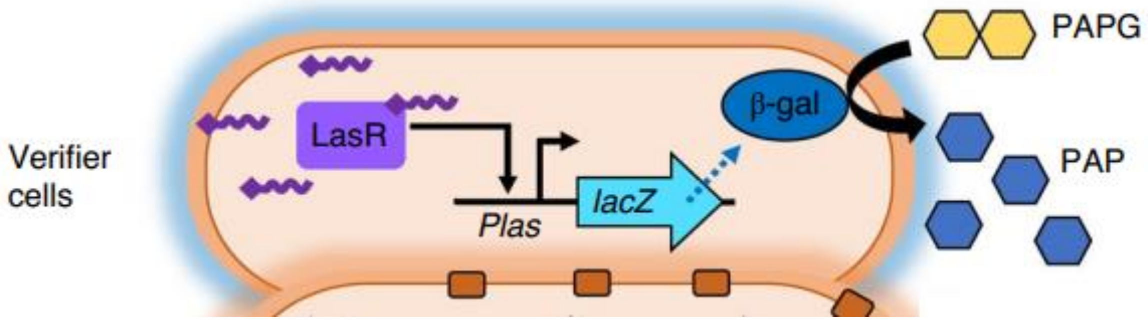
Step 3B: Verification signal activation

- Verifier cells are also induced by AHL in an identical manner to bioactuator cells via LasR-Plas binding.
  - Activates the *lacZ* gene and thus the synthesis of β-galactosidase
  - β-gal cleaves PAPG to PAP which can be electrochemically detected



AHL = Signal protein
LasR = Transcriptional activator
LacZ = Gene of interest
Plas = Promotor
β-gal = β-galactosidase
PAPG = 4-aminophenyl-β-D-galactopyranoside
PAP = *p*-aminophenol

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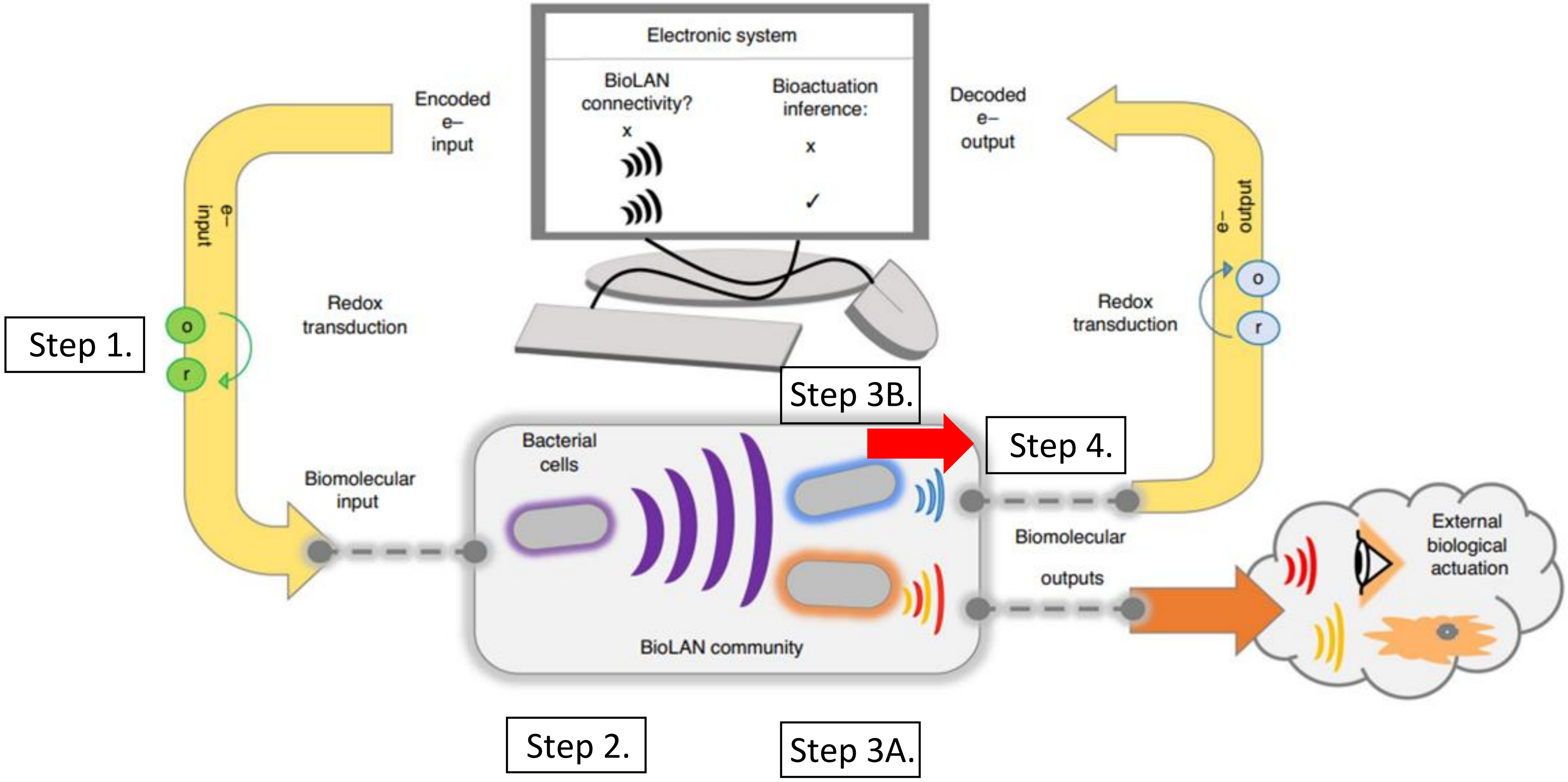
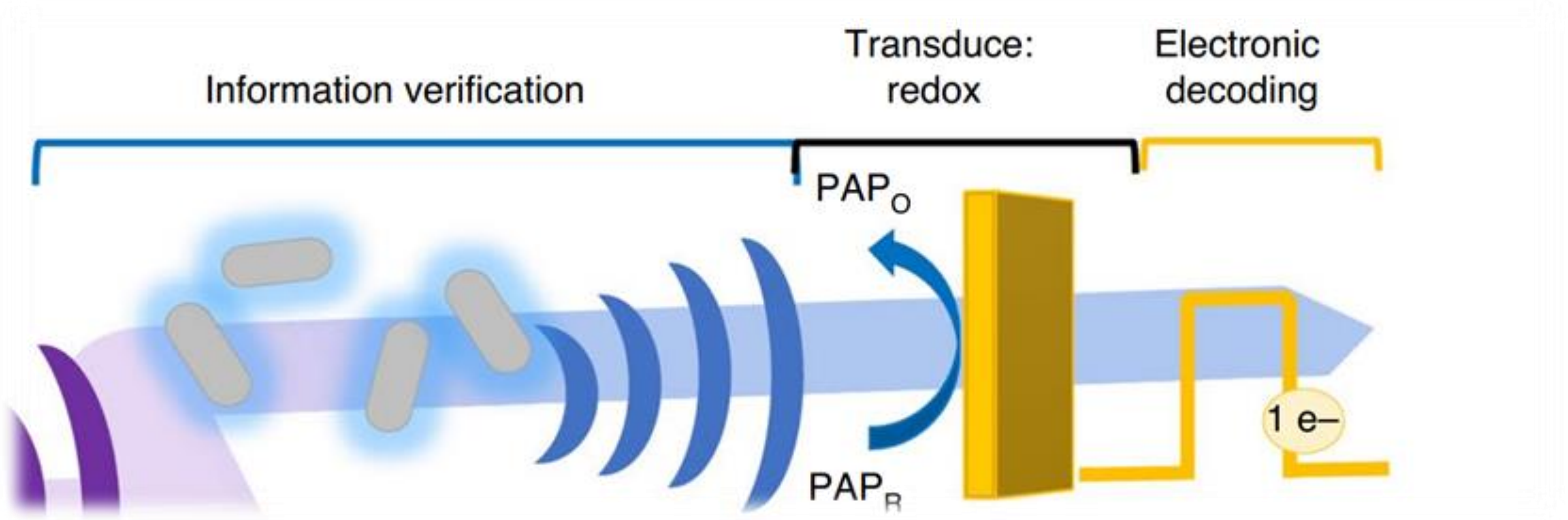

Electronic system
BioLAN connectivity?
Bioactuation inference:
x
x
✓
Encoded e– input
e– input
Decoded e– output
e– output
o
r
Redox transduction
Redox transduction
Step 1.
Biomolecular input
Bacterial cells
BioLAN community
Step 2.
Step 3A.
Step 3B.
Step 4.
Biomolecular outputs
External biological actuation

Step 4: Detection of verification signal

- PAP is oxidized which can be electrochemically detected
- The signal is propagated back to the electronic interface and decoded
- Confirms production of GMCSF (reflexive feedback)



PAP = *p*-aminophenol
e- = electron

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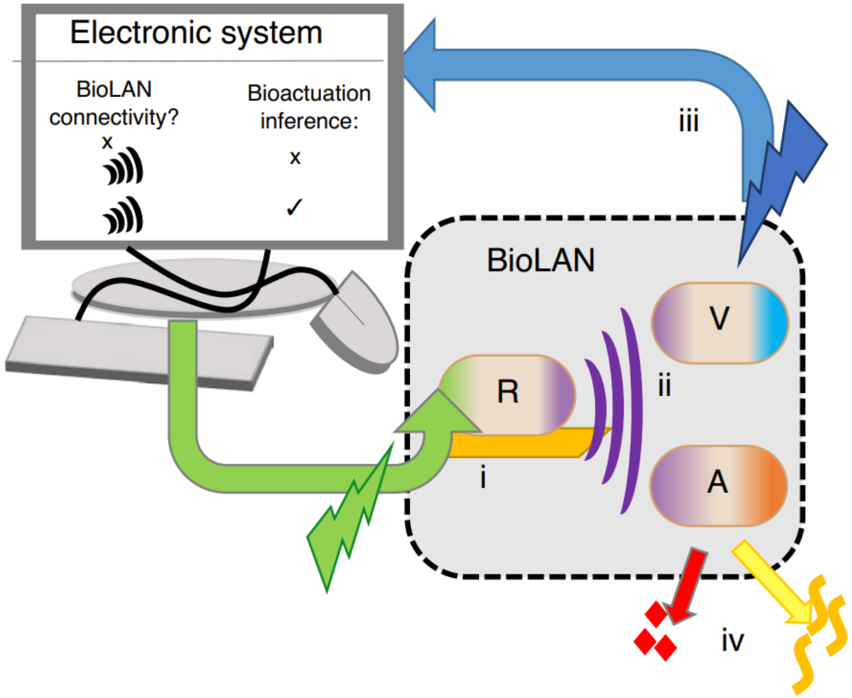
3. Discussion

What was achieved, future applications & Summary

What was achieved?

- Overall, the group were successful in creating a bidirectional bioelectronic system that drives programmed biological functions
- The target product was successfully produced, and production could be measured electronically.

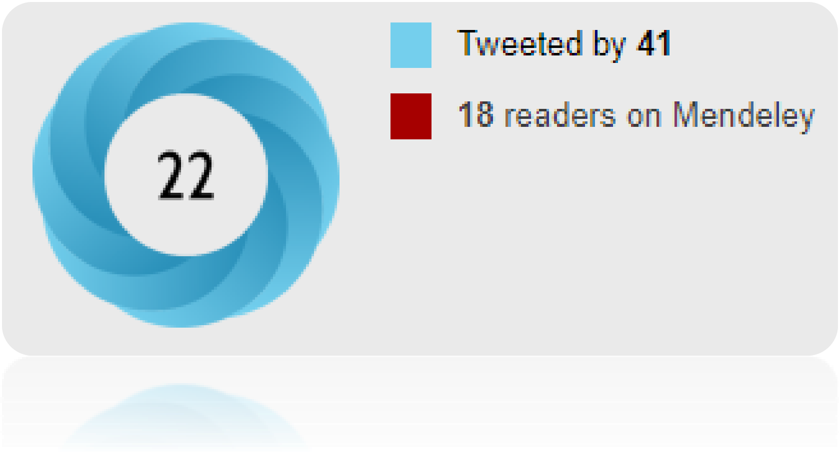


i = input charge
ii = AHL production levels
iii = electronic output from verifier cells
iv = production from actuator cells

R = router
V = verifier cells
A = actuator cells

Further Research

- Very recent article, published in March 2021
- Further optimization of parameters
- Experimentation with more complex systems both *in vitro* and *in vivo*
  - Use of logic gates
  - Future internet of Bio-Nano Things
  - Capability to embed biological intelligence
    - Ecological settings
    - Wearable interfaces
    - *In vivo* environments



Potential Applications

- Potential use for biohybrid devices
  - Electronically programmed functions can be signaled to living cells
- Ingestible capsules
- Environmental sensors
- Electronic tattoos
- Can be coupled with biological microelectromechanical system (Bio-MEMS) sensors and parts
  - Blood pressure sensors
  - Implantable microelectrodes – Interfacing with the body's nervous system
  - Microneedles for drug delivery

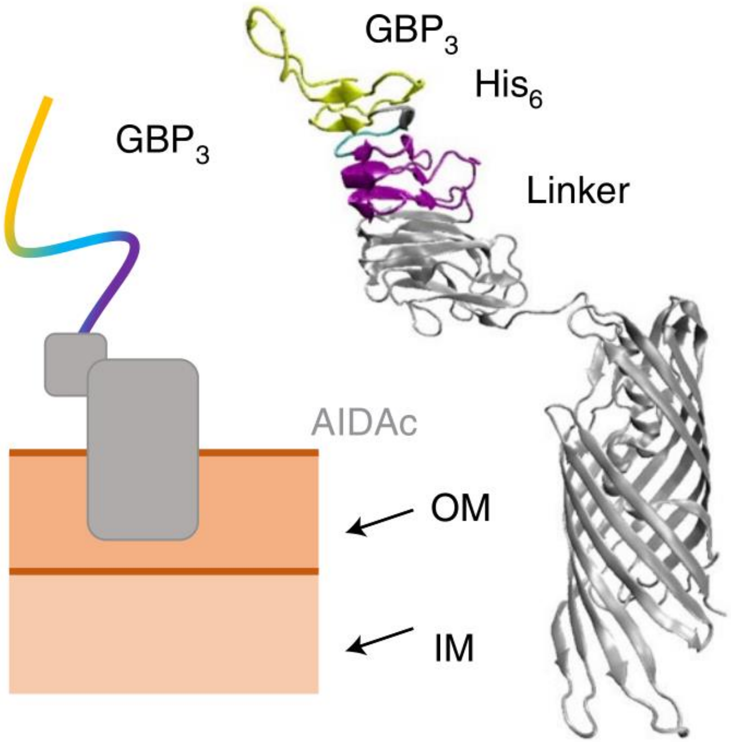
Summary

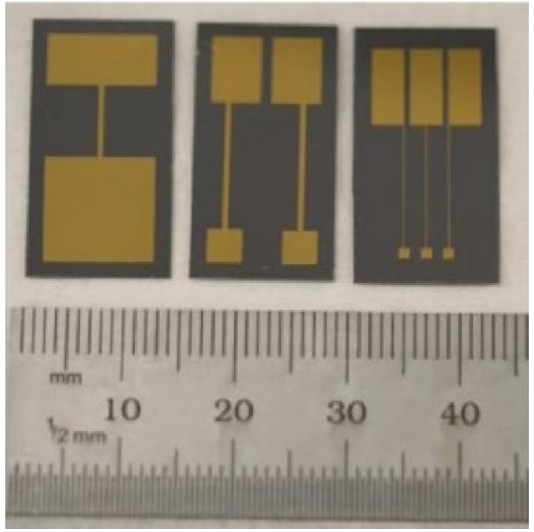
- BioLAN connects an electronic interface to biology and biology back to the electronic interface (Reflexive feedback)
- Because redox reactions are ubiquitous in biology, this has the potential to connect biology to a variety of electronic devices
- Allows for confirmation of the production of hard-to-detect molecules such as GMCSF

Thank you!

Questions?

Appendix





AIDAc = Autotransporter pore-forming protein
His_6 = Histidine
GBP_3 = Gold-binding peptide
OM = outer membrane
IM = inner membrane