**TEAM:** Elisha Krauss, Maddison Gahagan, Eric Grewal, Ellis Kelly, Abigael Schonewille, Emily Rae, Joel Tod, Julia Grein, Janis Cheng, Jimmy Chung, James Colwell, Alice Park, Alexander Pipchuk, Braedan Robinson, Daniel Stret, Sara Stickley, Declan Rowett, Ruben Warkentin
**ADVISORS:** Dr. John Allingham, Dr. Martin Petkovich, Dr. Virginia Walker, Dr. Robert Campbell

Queen's Canada presents

# IN THE GLOW

## *Luminescent Biosensors for Hormone Detection and Diagnosis*

opentrons
ThermoFisher SCIENTIFIC
Queen's University
Summer Work Experience Program
Dunin-Deshpande Queen's Innovation Centre
Department of Biomedical & Molecular Sciences
Office of the Associate Dean of Life Sciences & Biochemistry
Office of the Vice-Principal (Research)
Faculty of Arts & Science
Division of Student Affairs
Principal's Student Initiative Fund

## SUMMARY

Biomarkers, measurable indicators of a biological state, are routinely collected in clinical practice. Queen's Canada iGEM has sought to develop an innovative design to measures salivary hormones in infants, starting with cortisol. Our *Soothe Sayer* device is a pacifier equipped with a bioreceptor to monitor hormone levels and transmit to a smartphone over real time. The data, collected via an infant-friendly device, can be used toward non-invasive diagnoses, treatment monitoring, and studying hormone-related disorders in infants.

## POLICY & PRACTICES

**ENGAGEMENT & EDUCATION**
Science Rendezvous ● Canadian Undergraduate Technology Conference (CUTC) ● Science Quest ● SynBio Club

**COLLABORATIONS**
Ontario Genetically Engineered Machine Network (oGEM) ● Team Stony Brook ●Team Calgary ● Team Makerere University ● Team Toronto ● Queen's Biomedical Innovation Team (QBiT) ● Queen's Reduced Gravity Experimental Design Team (QRGX)


Science Rendezvous, Kingston, ON


Directed evolution video (Stony Brook × Queen's)


Science Quest, Queen's University

## BACKGROUND & MOTIVATION

Endocrine system responsible for various homeostatic controls and behaviours

Cortisol hormone is associated with stress ("fight or flight")

Cushing's syndrome and Addison's disease are caused by hyper- and hyposecretion of cortisol, respectively

Non-invasive salivary method gaining interest over blood tests

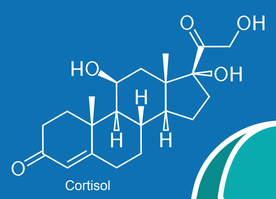
### Expert Interviews

*Interviewed 7 specialists/organizations of unique perspectives to guide device development (critical considerations below):*

- Time-efficient and affordable
- Accurate and non-disruptive
- Baseline readings
- Research and clinical use
- Sensory sensitivity (taste, texture)
- Other relevant metabolites


Dr. Janet Menard, Dept. of Psychology, Queen's University, Kingston, ON

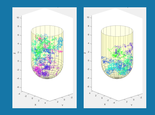

Dr. Kimberly Dow, Dept. of Pediatrics Queen's University, Kingston, ON



Cortisol

## MODELLING

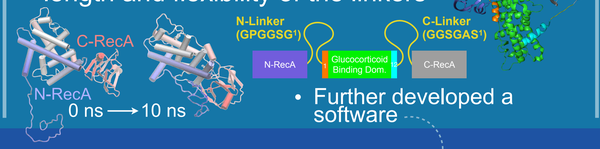
### Fluid Dynamics



- Saliva flow properties (viscosity, tension, Brownian motion)
- Movement (forces, capillary action)

### Molecular Dynamics & Linker Development

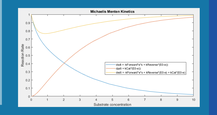
- Objective: To determine appropriate length and flexibility of the linkers




- Further developed a software

### Michaelis-Menten

- NanoLuc® Luciferase kinetics




## SMARTPHONE APP

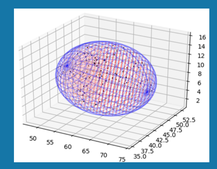
## SOFTWARE

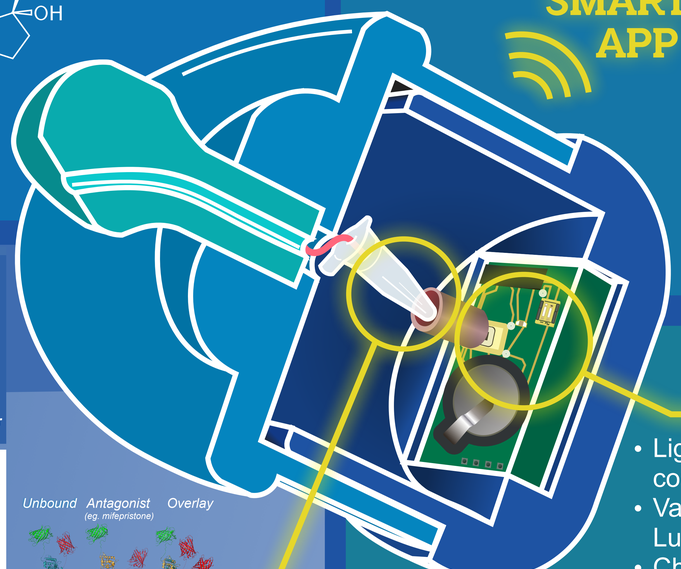
### Linker Software

- Connects two selected points on a molecule by finding the shortest path without interference (extension of Dijkstra's Algorithm)

### Luminometer Software

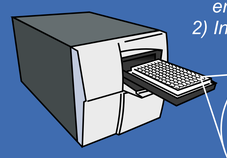
- Bluetooth-enabled (BLE) transmission


Ellipsoid points algorithm makes a grid of possible connection points



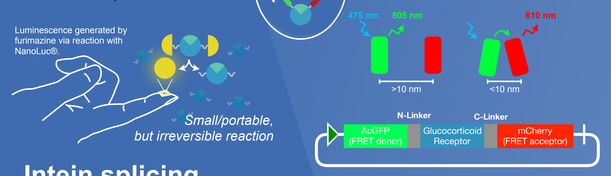
## DESIGN & LABORATORY

*Two design approaches: 1) Fluorescence resonance energy transfer (FRET) 2) Intein splicing*

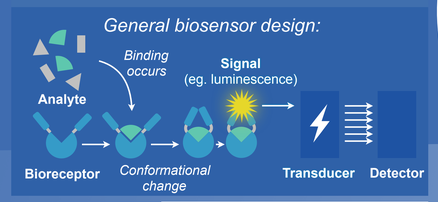


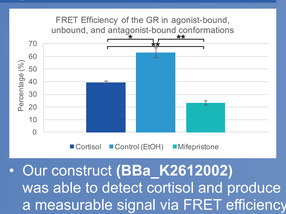
### FRET

*Reagentless, but not portable*

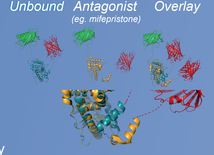



*General biosensor design:*



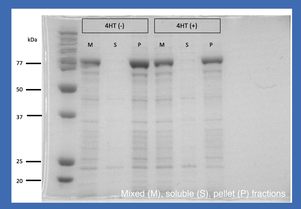

- Our construct **(BBa_K2612002)** was able to detect cortisol and produce a measurable signal via FRET efficiency




### Intein splicing

- Explored small-molecule triggered inteins[1] as customizable biosensors, with modular ligand binding domains, and exteins
- Used *Mycobacterium tuberculosis* RecA intein, with estrogen and glucocorticoid receptor ligand binding domains
- Piloted with KanR and 4-HT-dependent splicing[1], by growth on Kan plates **(BBa_K2612001)**

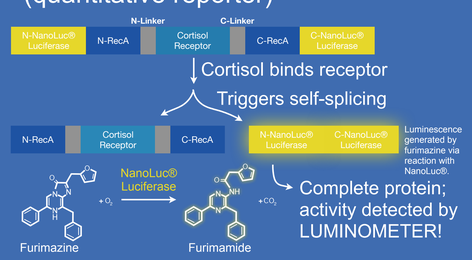

- NanoLuc® Luciferase coding device **(BBa_K2612000)**


- Our intern construct **(BBa_K2612001)** was insoluble in *E. coli* BL21 DE3, and accordingly, unable to splice

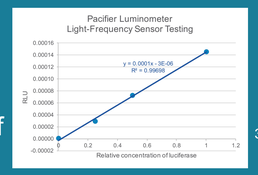
## BIOSENSOR

- NanoLuc® Luciferase as extein (quantitative reporter)

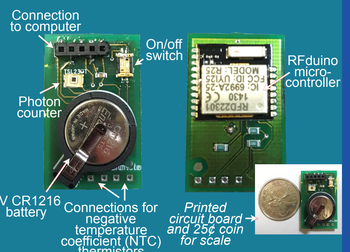



## LUMINOMETER

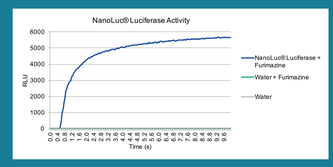
- Light frequency sensor counts photons over time
- Validation of NanoLuc® Luciferase for application
- Characterized sensitivity of our portable luminometer



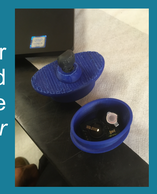

## HARDWARE

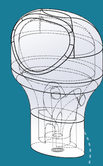


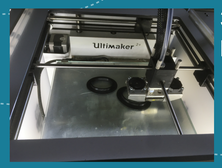

## PACIFIER DESIGN


Luminometer integrated into the *Soothe Sayer*




"Orthodontic" **nipple**



3D printing


**Saliva valve** is one-way to prevent backflow

**Saliva channel** designed with large channels near the mouth to maximize quantity and rate of collection; passively collects saliva by capillary action

*Special acknowledgment:*
QBiT
Queen's Biomedical Innovation Team

QGEM
Queen's Genetically Engineered Machine
@iGEMQueens
QueensiGEM.ca

**REFERENCE:** [1] Buskirk, A. R., Ong, Y.-C., Gartner, Z. J., & Liu, D. R. (2004). Directed evolution of ligand dependence: Small-molecule-activated protein splicing. *Proceedings of the National Academy of Sciences of the United States of America, 101*(29), 10505–10510.