Designing the LIT bulb

Step 1

Determine the ratio of each cell type for the co-culture in the LIT lightbulb

We calculated the number of *E. coli* cells required in the lightbulb to produce the same luminescence as a 160W lightbulb:

- Each *E.coli* produces a luminescence of $10^4 \frac{photons}{s}$
- A 160W lightbulb produces a luminescence of $10^{18} \frac{photons}{s}$

$$\frac{10^{18} \frac{photons}{s}}{10^4 \frac{photons}{s}} = 10^{14}\ E.coli\ cells\ are\ required\ to\ reach\ the\ desired\ luminescence$$

Therefore, 10^{14} *E.coli* cells are needed to produce the luminescence of a 160W lightbulb.

We then calculated the number of *Cyanobacteria* cells required in our LIT lightbulb:

- Source [1] conducts research on the co-culture of *Cyanobacteria* and *Yeast*. We assumed *Yeast* and *E.coli* take up the same amount of sugar for cell maintenance, therefore the same ratio of *Yeast* cells and *E.coli* cells will be used in their respective cultures with *Cyanobacteria cells*
- The ratio of *Yeast* cells to *Cyanobacteria* cells suggested in the paper was:
  - $10^7\ Yeast\ cell\ number : 10^8\ Cyanobacteria\ cell\ number$
  - $10^7\ E.coli\ cell\ number : 10^8\ Cyanobacteria\ cell\ number$
  - $\frac{10^7}{10^8} = \frac{10^{14}}{x}$
    $x = 10^{15}\ Cyanobacteria\ cells\ required\ in\ our\ lightbulb$

Therefore, we require 10^{15} *Cyanobacteria* cells to provide sufficient Oxygen and Glucose for the *E.coli* cells in the culture to survive.

Step 2

Determine the working volume of the LIT lightbulb

We calculated the volume of media needed for the co-culture of our two cell types.

Media requirements for the *Cyanobacteria* cells:

- The concentration of *Cyanobacteria* cells cultured per mL of media is:
  $$50{,}000\ \frac{\text{cells}}{\text{mL of media}} \times \frac{1{,}000\ \text{mL}}{\text{L}} = 50{,}000{,}000\ \frac{\text{cells}}{\text{L of media}}$$

  Therefore, since the total number of *Cyanobacteria* cells in our lightbulb is $1{\times}10^{15}\ cells$, we were able to determine the volume of media needed:
  $$\frac{1{\times}10^{15}\ cells}{50{,}000{,}000\ \frac{cells}{L\ of\ media}} = 0.002\ L \text{ of media required for the } Cyanobacteria\ cells$$

- Volume occupied by the *Cyanobacteria* cells:
  $$(1{\times}10^5\ cells) \times 0.2\ \mu m^3 = 20{,}000\ \mu m^3 \times \frac{1\ m^3}{(1{\times}10^6)^3 \mu m^3} = 2{\times}10^{-14}\ m^3 \text{ volume of } Cyanobacteria\ cells$$

Media requirements for the *E. coli* cells:

- The inoculum concentration conventionally used for *E.coli* cell fermentations is 5% w/w (cells to media)
- The total number of *E.coli* cells present in our lightbulb, $1\times10^{14}\ cells$, and the total volume of each cell, $0.7\ \mu m^3$, are both known. Therefore, the total volume of the cells in the lightbulb is:

  $(1\times10^{14}\ cells)\times0.7\mu m^3 = 7\times10^{13}\mu m^3$ volume of $E.coli$ cells

  $7\times10^{13}\ \mu m^3\times\frac{m^3}{(1\times10^6)^3\mu m^3} = 7\times10^{-5}\ m^3$ volume of $E.coli$ cells

- Given that the cells represent 5% of the total culture volume, we used the cell volumes to determine the volume of media required for our cell culture:

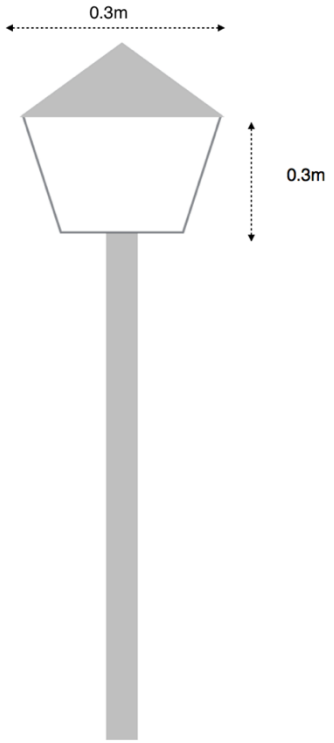
  $\frac{7\times10^{-5}\ m^3}{5}\times95 = 0.00133\ m^3\times\frac{1{,}000\ L}{1\ m^3} = 1.33\ L$ of media are required for the $E.coli\ cells$

We calculated the total volume of our lightbulb through adding the volume of each cell type and the respective volume of media they require:

$$2\times10^{-11}\ L\ + 0.002\ L + \ 0.07\ L + 1.33\ L = 1.402\ \cong\ 1.5\ L$$

Step 3

Determine the dimensions and properties of the conventional streetlamps in London



- Material of the glass shell: Plexiglas
- Assuming the shape of the streetlamps container for the lightbulb is a cylinder

  $V = \pi\times h\times r^2$
- Volume available for the bacterial co-culture

  $V = \pi\times0.3\times0.12^2$

  $V = 0.0136\ m^3$

Fig. 1 Sketch of a standard street lamp in London

Alternative designs considered for the LIT lightbulb

Idea 1- Bioreactor Model

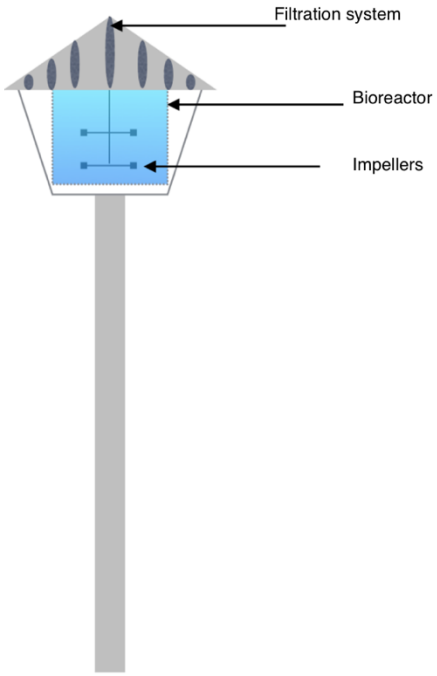


Fig. 1 Depicts the Bioreactor Model LIT bulb implemented in a streetlamp. A filtration system is in place to clean the air entering coming in contact with the bacterial cultures. The Bioreactor contains the cell co- culture and media. The lid of the reactor is made out of an oxygen permeable material, the impeller is comprised of 2 Rushton turbines to ensure homogeneity is maintained in the reactor.

Assumptions

- The shape of the reactor is a cylinder (Therefore, $\mathrm{V} = \pi \times h \times r^2$)
- The material of the bioreactor can: resist and maintain its permeability at extreme temperatures; withstand high shear forces; allow sunlight to pass through it as it is transparent; and complies with REACH requirements
- The working volume of the Bioreactor is 80%
- The aspect ratio, the ratio of the height of the bioreactor to the diameter of the bioreactor, (h_B:d_B), is 3:1
- The ratio of the diameter of the Bioreactor to the diameter of the impeller (d_B: d_i) is 3:1
- The existing electrical installation in the streetlamps can provide the energy required to power the rotation of the impeller
- Rushton turbines are used as bacterial cells are being handled, therefore they can withstand the shear forces exerted from the impellers while at the same time require large volumes of oxygen as they respire at a fast rate than mammalian cells
- 4 baffles are required to prevent the formation of vortexes from the rotation of the impeller

Issues identified with design:

- There is no control over the heat generated by mechanical rotation and cell respiration, therefore, to ensure the cells do not overheat a cooling jacket would need to be incorporated in the design
- There is no efficient way to monitor Oxygen levels, pH, and temperature within the vessel
- There is a high risk of bio contamination, as if the street lamp breaks, the content of the bioreactor would spill and get released into the environment

Idea 2- Cellular encapsulation in hydrogels

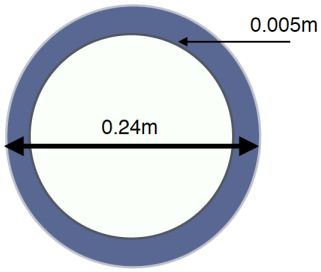


Fig. 3 Cross-sectional shot of the currently in place streetlamp in London

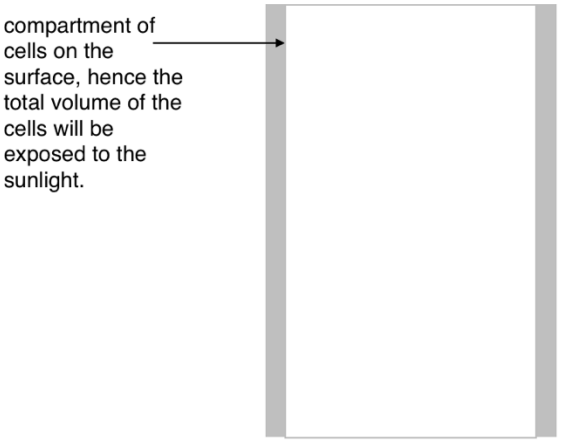


Fig. 4 Horizontal view of the cylinder streetlamp, displaying the area where the cell cultures will be placed.

Volume of the cell compartment:

$$V_C = V_T - V_I$$
$$V_C = (\pi \times h \times r^2) - (\pi \times h \times r^2)$$
$$V_C = (\pi \times 0.3 \times (0.120)^2) - (\pi \times 0.3 \times (115)^2)$$
$$V_C = 2.26 \times 10^{-3} m^3$$

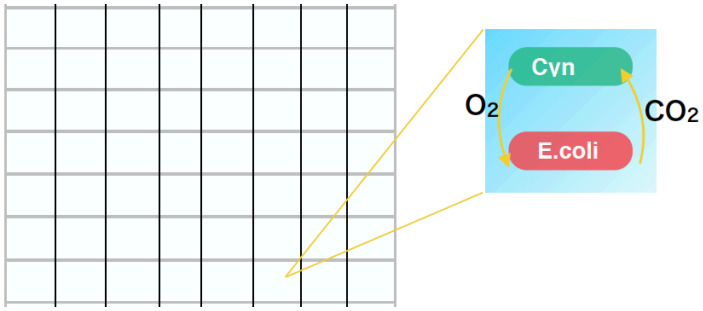


Fig. 5 Detailed view of the structure of the cell compartments, each compartment is made out of TPX™ and contains M9 media and cyanobacteria and E.coli co-culture

Properties of the material (**TPX™**) of the cell culture compartments:

- Chemical resistance to chemicals such as ethanol and acetone (both of which could be products from anaerobic respiration from the bacterial cells)
- Resistance to heat, hence the individual culture compartments will not degrade if exposed to sunlight
- Transparency, this allows for a high amount of sunlight to pass through the material and therefore increase the efficiency of the bulb

- Low reflective index
- High permeability to oxygen, which is required to encode the bioluminescence in the bacterial construct

Issues with the design:

- Homogeneity within each individual compartment cannot be maintained, therefore eventually the cells will sediment
- As the cells will line the glass there is risk the cells may become overheated and therefore could denature. This could be tackled through introducing a bio brick construct within the plasmid of the cells that gives them resistance to extreme temperatures
- There is no efficient way to monitor Oxygen levels, pH, and temperature within the vessel
- There is no control over the heat generated by mechanical rotation and cell respiration, therefore, to ensure the cells do not overheat a cooling jacket would need to be incorporated in the design

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