

Microscope Cell Culture Incubator

Preliminary Product Design Specifications

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Client: Dr. John Puccinelli

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Function:

To support live cell imaging in the BME teaching lab, the client needs a cell culture incubator that fits on the stage of a Nikon TI-U Inverted Fluorescence Microscope. The device must maintain constant environmental conditions (37°C, 100% Humidity, and 5% CO₂ concentration) for up to one week. Moreover, the incubator must have an optically clear top and bottom that do not interfere with fluorescence imaging. The device should accommodate different types of cell culture containers including multiwell plates, chamber slides, rectangular dishes, and Petri dishes. The client would like the device to display the environmental conditions and notify the user if conditions deviate from an acceptable range. Commercially available devices satisfy these design requirements, although they are expensive; the client's budget is \$100 per device.

Client Requirements:

- Maintain 37°C, 100% Humidity, and 5% CO₂ concentration
- Does not impede the optical path
- Allow cell plates with a maximum size of 130mm x 90mm x 20mm
- Uniform heating throughout the incubator
- Easy readout of conditions
- Ability to change out cell cultures
- Ability to be sterilized
- Combined budget: \$100

Design Requirements:

Performance Requirements

- Must be able to maintain uniform heat distribution, CO₂ concentration, and humidity within the incubator
- Limit the condensation within the incubator as to not visually impair the microscope

Safety

- Compliant with standards set by Biosafety Level 1
- Can controllably deliver CO₂ from a tank regulator maintained at 15 psi

Accuracy and Reliability

- Maintain a temperature of 37°C ± 1°C

- CO₂ concentration 5% ± 0.5%
- 95%-100% humidity

Life and Service

- The incubator must maintain these conditions for one week
- The electronics should not have to be adjusted or fixed during this time to preserve sterility

Shelf Life

- 3 years

Operating Environment

- Used within a Biosafety Level 1 environment
- All electronics must be sterile and water resistant
- System will have to adjust to changing environmental conditions due to opening and closing of the incubator

Ergonomics

- Device should be easily accessible for user
- Should be big enough for user to easily place and remove cell culture plate, flask, or petri dish
- Incubator door should be easy to open

Size

- Allow cell plates with a maximum size of 130mm x 90mm x 20mm
 - Slightly bigger than the size of a 96 well plate

Weight

- Lightweight as to allow the device to be easily movable
- Cannot be more than 5kg

Materials

- Transparent on the top and bottom for the microscope. Other incubator materials need to be heat resistant from the high humidity levels and sterilized

Aesthetics, Appearance, and Finish

- The materials through which imaging occurs needs to be transparent so as to not interfere with the image quality.
- There should be a user interface to display the current conditions of the incubator

Production Characteristics:

- *Quantity:* 1 design
- *Target Production Cost:* less than \$100

Miscellaneous:

Standards and Specifications

- None

End User Concerns

- Easy enough for undergraduate students to use

Competition

- EVOS Onstage Incubator [1]
 - Fully integrated environmental chamber for live cell time-lapse imaging
 - Easily maintain physiological conditions

- Sleek design
- Wide range of vessel holders
- Okolab Stage Top Incubator [2]
 - Temperature Accuracy: $\pm 0.1^{\circ}\text{C}$ in sample feedback mode, $\pm 0.3^{\circ}\text{C}$ in chamber feedback mode
 - Compatible with: Bold Line T, UNO Combined and H401-T Controllers
 - Embedded temperature sensor in heated glass lid and in chamber body
 - Interchangeable magnetic inserts allow to host Petri, Slides and MW Plates
 - Magnetic locks hold the Petri, the Slides and the MW Plates in the correct position inside the chamber
 - Perfusion holes available for inlet and outlet of tubes
 - Sliding lid allows easy access to the sample
- World Precision Instruments Stagetop Environmental Chamber with Controller [3]
 - Four programmable digital control loops:
 - Independent incubator base temperature PID control with $\pm 0.1^{\circ}\text{C}$ precision
 - Independent incubator lid temperature PID control with $\pm 0.1^{\circ}\text{C}$ precision
 - Airflow digital PID control from 0–900 SCCM
 - USB-based remote control and data logging
 - Electronic flow meter
 - Programmable alarm for out of tolerance condition on all four channels
 - Compact and lightweight
 - Monitor and control the flow and temperature in a microscope chamber or stagetop environment
 - \$11,500.00
- Ibidi Stage Top Incubation System [4]
 - Precise control of temperature, humidity, CO₂, and O₂
 - Compatible with all inverted microscopes that have a K-Frame stage (160 mm x 110 mm)
 - Prevents condensation by heating culture dish lid
 - \$13,990.00

Sources

- [1] Sciences, L., Analysis, C., Imaging, C., Systems, E. and Incubator, E., 2020. EVOS Onstage Incubator | Thermo Fisher Scientific - UK. [online] Thermofisher.com. Available at: <https://www.thermofisher.com/us/en/home/life-science/cell-analysis/cellular-imaging/cell-imaging-systems/evos-onstage-incubator.html?ef_id=Cj0KCQjwZ7BRDzARIsAGjbK2YL6vWkY6uDHxXWhmKWyn41Hm65MqJSE1mAdg6mG6yM69Hl1kHxLfkaAkKNEALw_wcB:G:s&s_kwid=AL13652!3!429011940942!b!!g!!%2Bmicroscope%20%2Bincubator&cid=bid_pca_iie_r01_co_cp1359_pjt0000_bid00000_0se_gaw_nt_lgn_ins&gclid=Cj0KCQjwZ7BRDzARIsAGjbK2YL6vWkY6uDHxXWhmKWyn41Hm65MqJSE1mAdg6mG6yM69Hl1kHxLfkaAkKNEALw_wcB> [Accessed 16 September 2020].
- [2] S. User, "Okolab Stage Top Incubator," Okolab - Stage Top - Digital Gas. [Online]. Available: <http://www.oko-lab.com/live-cell-imaging/stage-top-digital-gas>. [Accessed: 17-Sep-2020].
- [3] Wpiinc.com. 2020. *Stagetop Environmental Chamber With Controller For Heat, Air Flow From Bottled Gas*. [online] Available at: <<https://www.wpiinc.com/st-h5-iv-stagetop-environmental-chamber-with-controller-for-heat-air-flow-from-bottled-gas>> [Accessed 18 September 2020].
- [4] ibidi. 2020. *Ibidi Stage Top Incubator | O2 & CO2 Regulation | K-Frame | Live Cell Imaging*. [online] Available at: <<https://ibidi.com/stage-top-incubators/233-ibidi-stage-top-incubation-system-multi-well-plates-k-frame-co2o2.html>> [Accessed 18 September 2020].