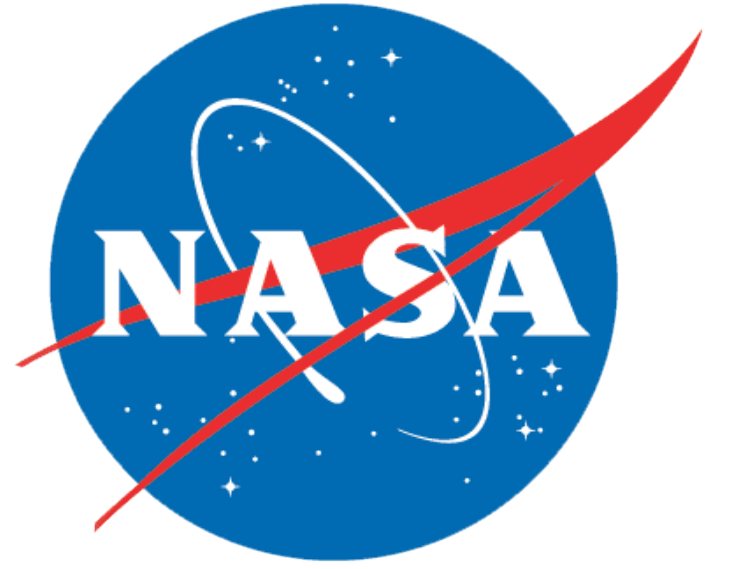




Active Control of pH in the Bioculture System through Carbon Dioxide Control

National Aeronautics and Space Administration



Luke Monhollon¹, David Pletcher², and Jessica Hauss²

¹Space Life Sciences Training Program (SLSTP), NASA Ames Research Center, ²Space Biosciences Division, NASA Ames Research Center

ABSTRACT

For successful cell research, the growth culture environment must be tightly controlled. Deviance from the optimal conditions will mask the desired variable being analyzed or lead to inconsistencies in the results. In standard laboratories, technology and procedures are readily available for the reliable control of variables such as temperature, pH, nutrient loading, and dissolved gases. Due to the nature of spaceflight, and the inherent constraints to engineering designs, these same elements become a challenge to maintain at stable values by both automated and manual approaches. Launch mass, volume, and power usage create significant constraints to cell culture systems; nonetheless, innovative solutions for active environmental controls are available. The acidity of the growth media cannot be measured through standard probes due to the degradation of electrodes and reliance on indicators for chromatography. Alternatively, carbon dioxide sensors are capable of monitoring the pH by leveraging the relationship between the partial pressure of carbon dioxide and carbonic acid in solution across a membrane. In microgravity cell growth systems, the gas delivery system can be used to actively maintain the media at the proper acidity by maintaining a suitable gas mixture around permeable tubing. Through this method, launch mass and volume are significantly reduced through the efficient use of the limited gas supply in orbit.

EXPERIMENTAL AIMS

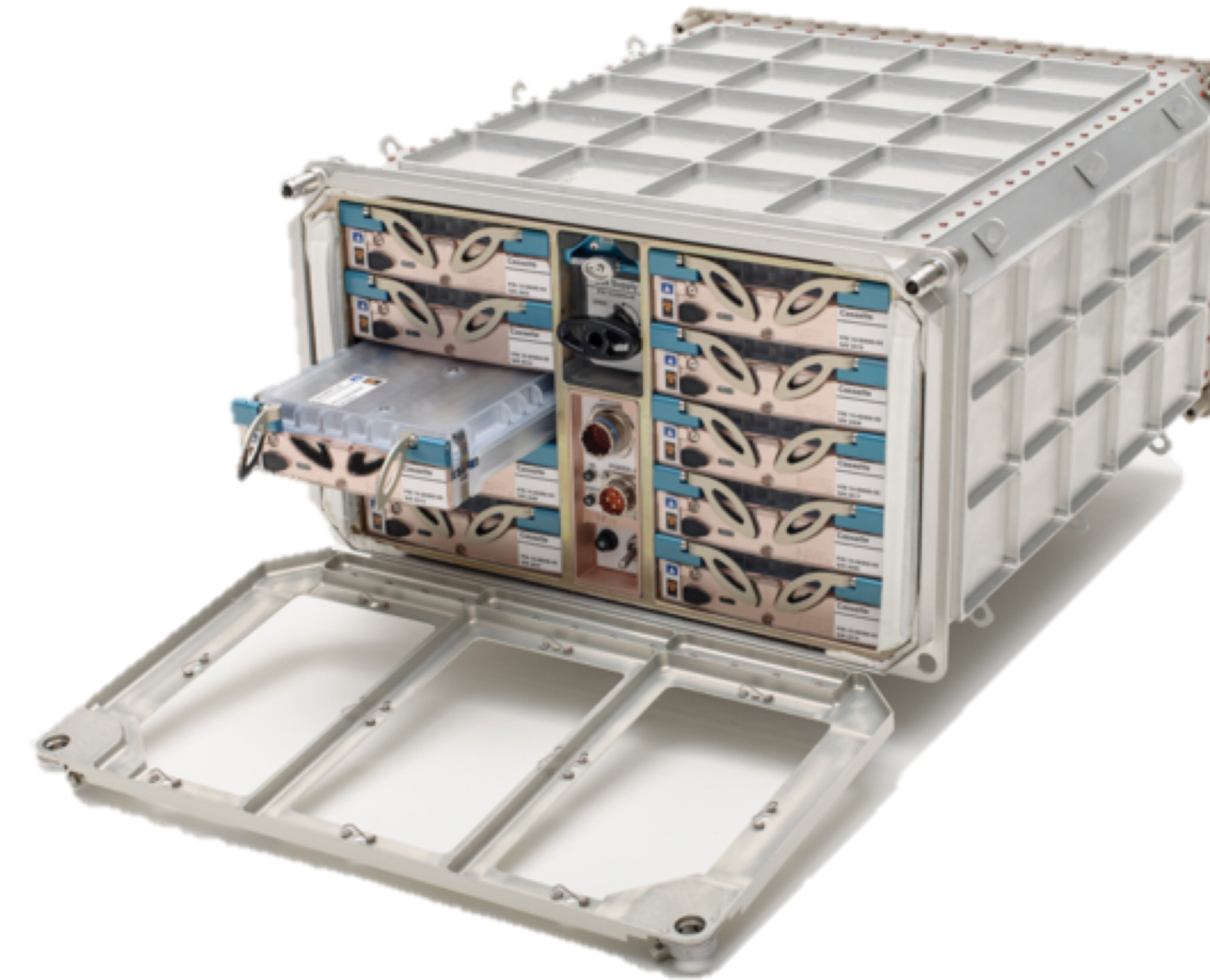
- (1) Characterize pump performance to confirm expected media delivery to bioreactor in different orientations
- (2) Determine gas dose delivery volume with respect to time
- (3) Analyze the relationship between CO₂ concentration and the media acidity response to optimize pH control

METHODS

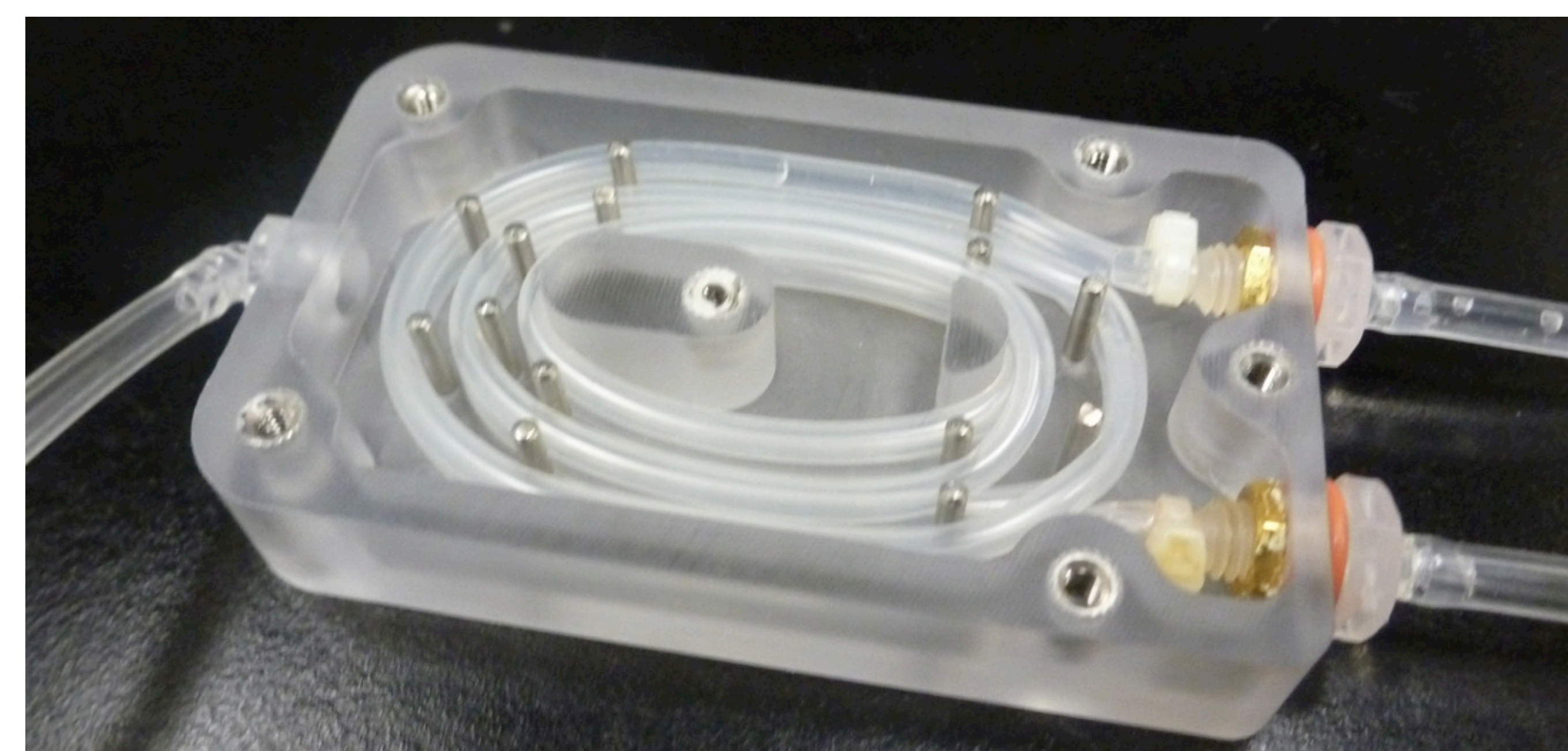
- (1) Collect volume flow rate measurements while anchoring the cassette and flow path in 5 different orientations, with and without the bioreactor installed
- (2) Measure volume of gas released from the docking station's 10 valves during actuations times from 80 to 300 milliseconds
- (3) Modulate CO₂ delivery while monitoring media pH and CO₂ concentrations in the oxygenator and inside the cassette

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Bioculture System with all cassettes installed



BACKGROUND

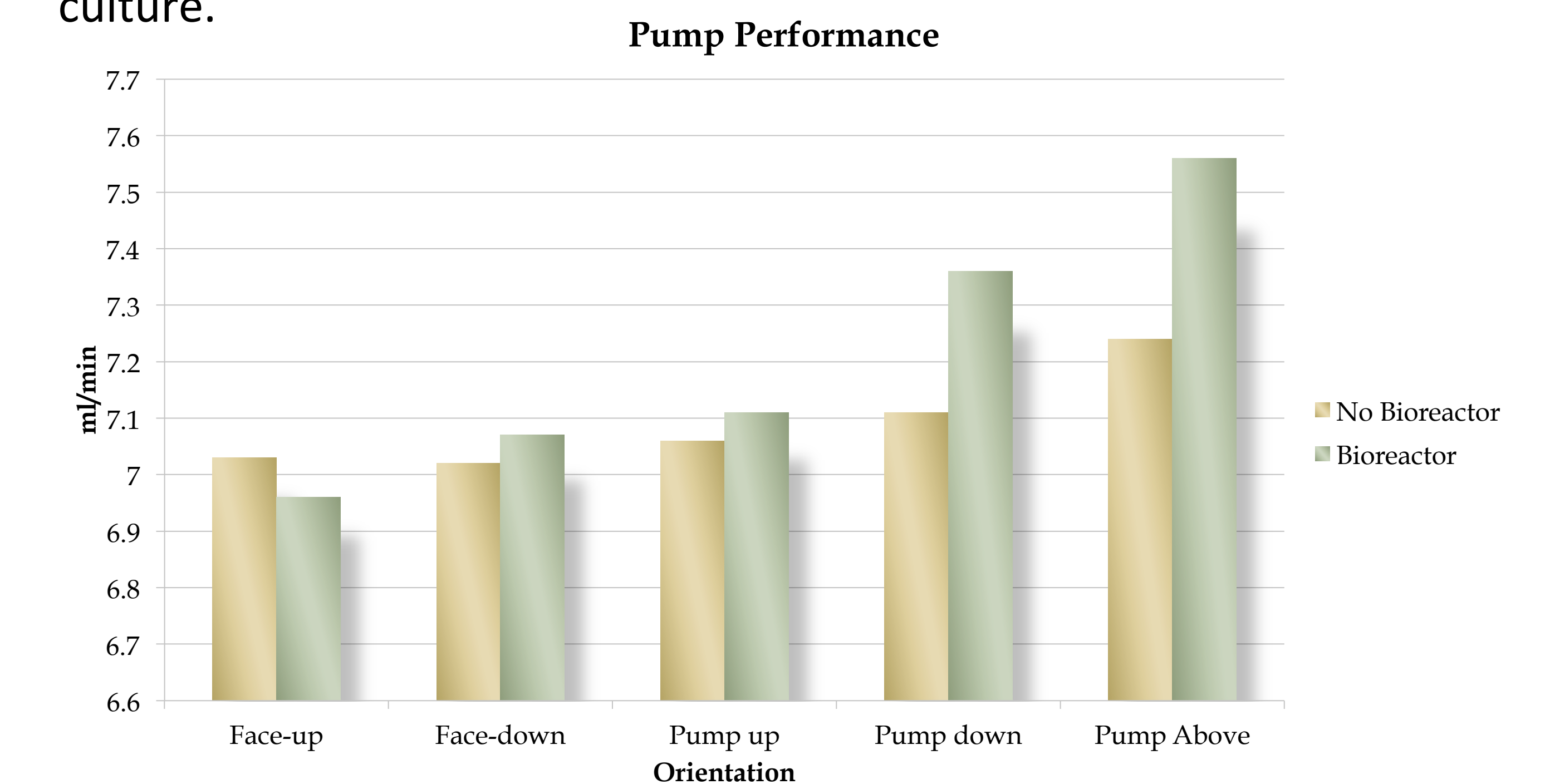


Flow path Media Oxygenator, lid removed.

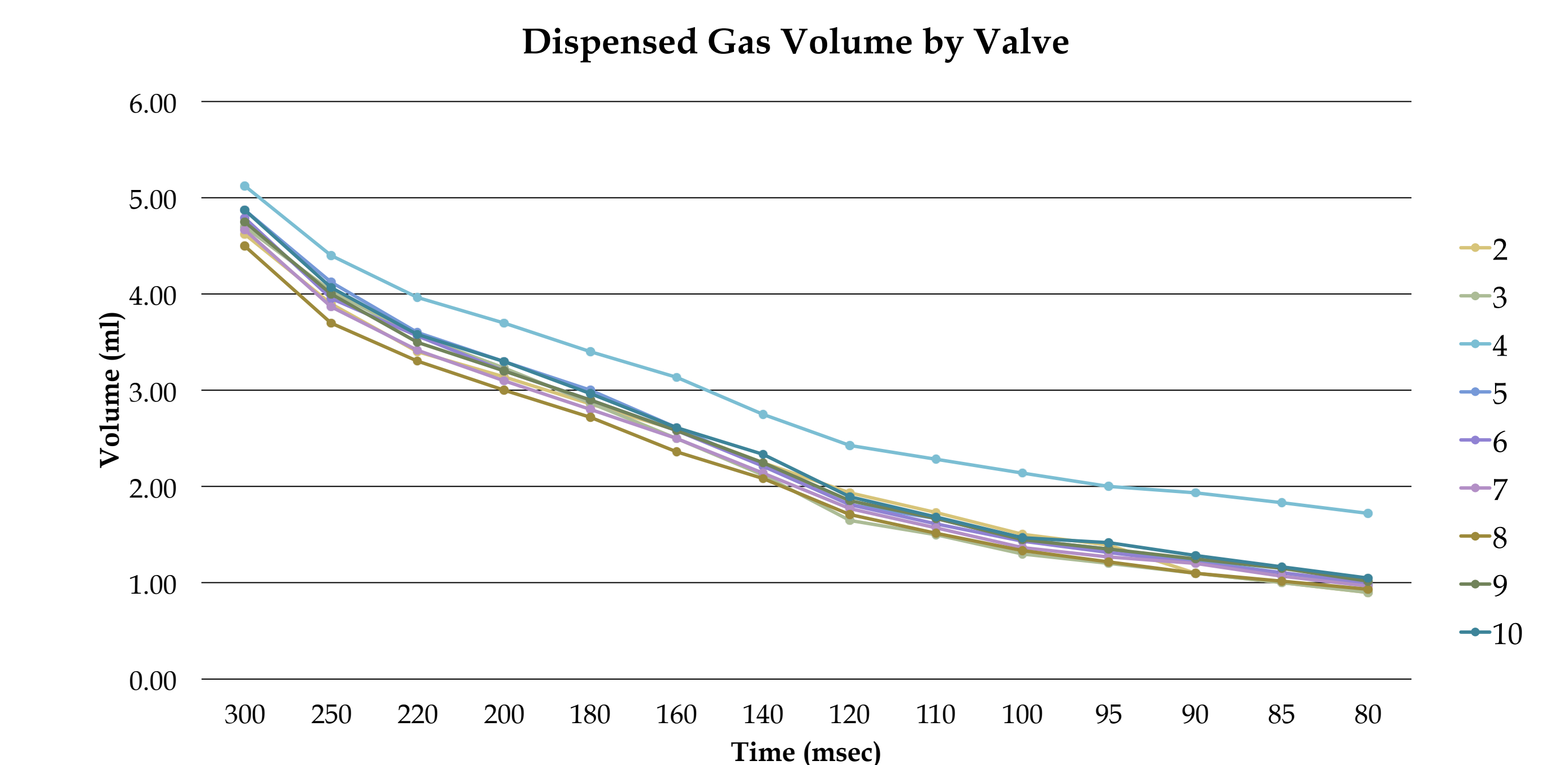
- Buffering system for media is carbonic acid/carbonate based.
- Maintenance is done by addition of gas (10% CO₂) to flow path.
- Gas/liquid diffusion is done through silicon tubing within oxygenator
 - Secondary diffusion occurs within the sealed cassette through flow path tubing.
- Cell culture is done within a hollow-fiber bioreactor.
 - Permeable fibers shield cells from mechanical stimulus
- Exchanges of media happen periodically by pumping used media to a sump bag and extracting fresh media from a storage bag.
 - Sump and media bags are refrigerated
 - Media is preheated in separate reservoir before coming into contact with the bioreactor.

RESULTS: Volume Flowrate

The media flow rate was within the expected range for proper cell culture.



RESULTS: Gas Delivery



RESULTS: pH Response to CO₂

The acidity was successfully controlled between 7.2-7.6 pH. As expected, the dose volume and rate had to be modulated to maintain the system. Due to the increased cell yield in the system, the correlation of the pH and partial pressure of CO₂ began to deviate as metabolites and cellular respiration rose. More research is needed to explore this effect for long term cell growth.

ACKNOWLEDGEMENTS

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