Integration of a robotic small scale bioreactor system as a prerequisite for a self-learning & autonomous cultivation platform

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Motivation / Introduction

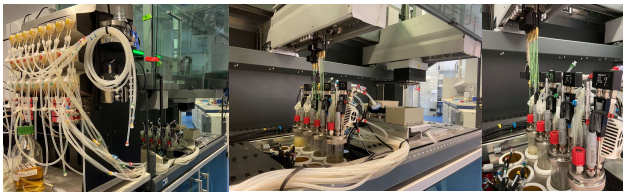
The process industries are moving towards digitalization and automation, known as the industry 4.0. In contrast to that, the biotechnological sector lacks this transition and does not even fulfill the automation standards of industry 3.0, while facing the necessity of faster development cycles [1]. In order to enhance automated high-throughput process development (HTBD) and optimization under consistent scalable conditions, we present the integration of a commercially available small scale bioreactor system, a liquid handling system (LHS) & a microplate reader into our HTBD-robotic facilty [2]. The integration and data management follows the F.A.I.R data principle [3], storing the corresponding on-line, at-line & off-line measurements of each bioreactor, as well as the executed liquid handling steps, in a centralized database, enabling further deployment. An initial fed-batch cultivation with automated sampling is used for verification of the conceptual design of the facility.

Concept and Results

BioXplorer Facility

8x parallel cultivation | **Automated sample handling** | ***At-line* analysis**

- **Cultivation system HEL BioXplorer 100**
- **LHS Tecan Evo 150**
- **Microplate reader**



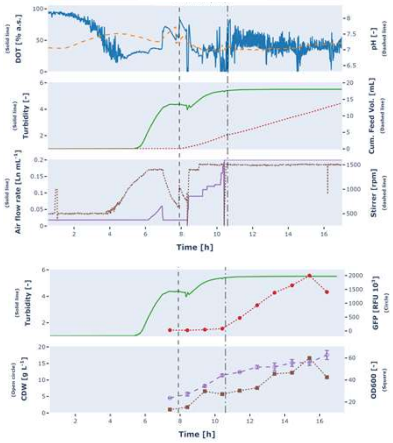
Cultivation system
- 8 vessels à 25 -150 mL, 1 MFC per vessel , 3 peristaltic pumps per vessel, batch, fed-batch & continuous mode

Monitoring *on-line*
- pH, DO, turbidity, temperature, stirring speed, feed rate, air flow rate,

Monitoring *at-line*
- automated sampling for substrate, metabolites, product, biomass,

Individual process control
- pH, DO, temperature, stirring speed, air flow rate, pump rates, inducer addition,

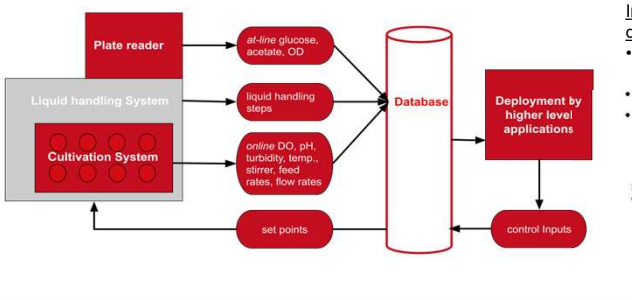
Proof of Concept



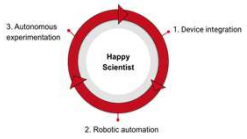
Figures Upper: *Online* process data for DOT, pH, turbidity, cumulated feed volume, air flow rate & stirrer speed. Lower: *Online,* & *offline* data for biomass & *at-line* data for fluorescene of GFP. First vertival dashed line indicates feed start & second vertical dashed/dotted line indicates induction.

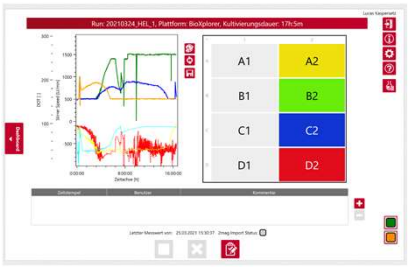
E. coli BL21 (DE3) with GFP-fusion (pET28 based reporter system) as protein of interest cultivated in fed-batch process (batch, exponential phase followed by induction phase). *Online* process data was logged every 30s, while *at-line* samples for OD_{600} and fluorescence's (479 nm excitation, 520 nm emission) and *off-line* samples for CDW were taken every hour. *On-line* turbidity measurements were limited at higher higher cells densities > 12 g L^{-1} and complemented through *at-line* OD_{600} and *offline* CDW measurements. Additionally, fluorescence measurements allow for product quantification, Hence, valuables process insights can be gained.

Integration into HTBD facility



Integration into centralized database
- *on-line* & *at-line* data submission of all devices
- reading external set points
- prerequisite for further deployment/ automation steps





Live data in centralized database allows for further automation steps, data driven process development and deployment by modelling frameworks and artificial intellegence based tools developed in the KIWI-biolab.

Conclusions and Outlook

I. Successfully integration of H.E.L BioXplorer into LHS for automated sampling procedure
II. Proof of concept with an industrial relevant fed-batch cultivation
III. Additional process insight through *at-line off-line* process data enabling data driven real-time optimization
IV. Connection to centralized database as a prerequiste for furhter automation steps deployment by AI based tools
V. Finalize integration steps for remote setpoint writing, and logging of LHS steps
VI. Further automate sampling and metabolite analysis by integration of a mobile lab assistant into the HTDB facility

Acknowledgements / References

This work was supported by the German Federal Ministry of Education and Research through the Program "International Future Labs for Artificial Intelligence" (Grant number 01DD20002A).
We gratefully thank Dr. Stefan Schiller for providing the strain.

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SPONSORED BY THE Federal Ministry of Education and Research