

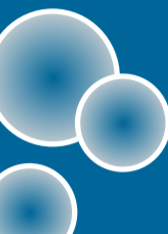
# Impact of initial glucose concentration on oxygen uptake rate in cell culture processes

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## Objective

In this work, the impact of the initial glucose concentration on cellular metabolic activity was investigated using the specific oxygen uptake rate ( $q_{OUR}$ ) and specific lactate uptake rate ( $q_{Lac}$ ) as indicators. To calculate  $q_{OUR}$ , reliable off-gas measurements are obtained using BlueSens off-gas analyser.



## Measurement principle for $q_{OUR}$

A global mass balance was applied to calculate  $q_{OUR}$  using the oxygen fractions ( $y_{O_2}$ ) in the incoming and outgoing gas streams. Using the BlueVary (BlueSens) variable off-gas analyser, the composition of the off-gas from the cell culture bioreactor was quantified. The workflow and basic calculations are shown in Figure 1.

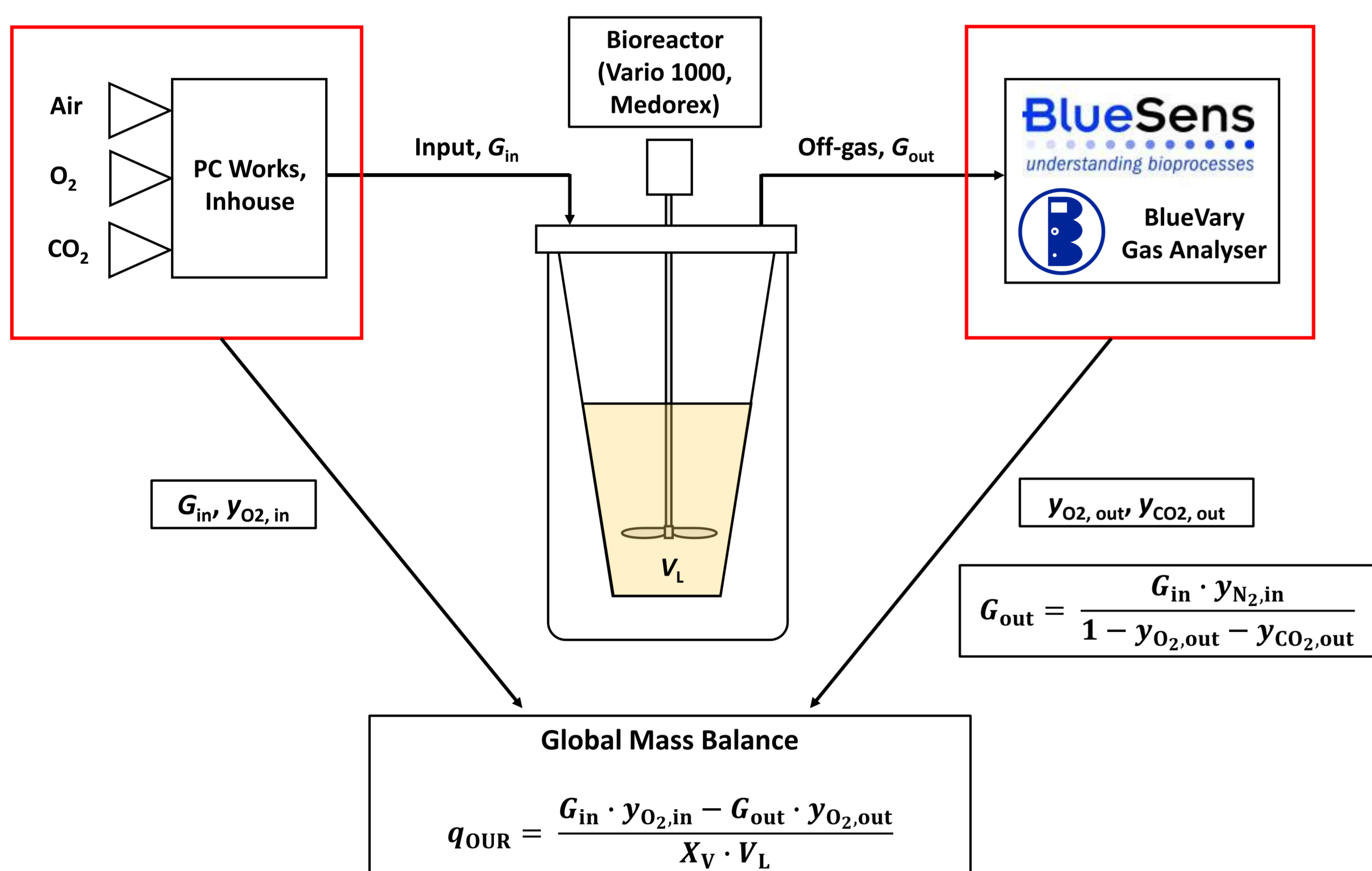


Figure 1: Workflow for calculating  $q_{OUR}$  using gas analyser

$y_{O_2}$	Volume fraction of oxygen in a gas stream
$G$	Volumetric gas flow rate
$X_V$	Viable cell density
$V_L$	Liquid working volume in the bioreactor



## Results and Discussion

The impact of two different initial glucose concentrations on the cell metabolic activity was investigated using off-gas measurements for  $q_{OUR}$  quantification. Additionally, specific growth rate during exponential phase ( $\mu_{exp}$ ), specific lactate uptake rate ( $q_{Lac}$ ) and specific antibody productivity ( $q_{mAb}$ ) were compared.

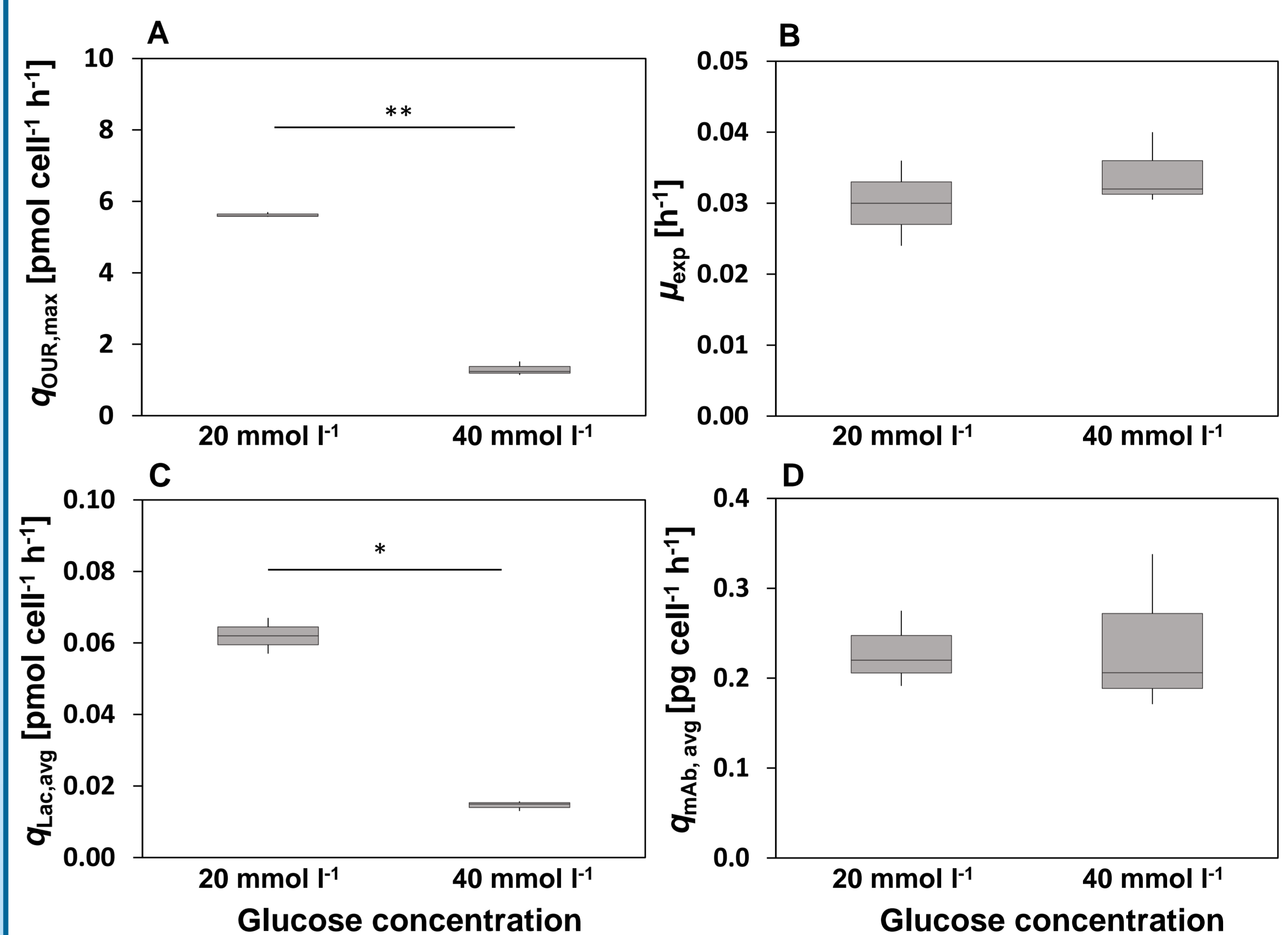


Figure 2: Comparison of specific rates for CHO batch cultivations with 20 mmol l<sup>-1</sup> and 40 mmol l<sup>-1</sup> initial glucose concentrations, A: Maximal specific oxygen uptake rate B: Specific growth rate (0 – 72 h), C: Specific lactate uptake rate during lactate consumption (averaged, t > 90 h) D: Specific productivity (averaged for all measurements), Working volume: 200 ml, Temperature: 37°C, Agitation: 400 – 800 rpm. Two-tailed independent sample t-test: \* = significant difference ( $p < 0.05$ ), \*\* = highly significant difference ( $p < 0.001$ )

Fig. 2A shows that the culture with 20 mmol l<sup>-1</sup> initial glucose culture has a four-fold higher  $q_{OUR,max} = 5.6$  pmol cell<sup>-1</sup> h<sup>-1</sup>, indicating a higher metabolic activity. This is not apparent from the similar growth rates (Fig. 2B) but can be inferred only through off-gas measurements and  $q_{OUR}$  calculations. The cells grown with 20 mmol l<sup>-1</sup> initial glucose also take up lactate more actively (Fig. 2C,  $q_{Lac,avg} = 0.06$  pmol cell<sup>-1</sup> h<sup>-1</sup>), which can help reduce the deleterious effects of high lactate levels [1]. Fig. 2D shows an average  $q_{mAb,avg} = 0.23$  pg cell<sup>-1</sup> h<sup>-1</sup> for both the initial glucose concentrations, which is also comparable to previous results [2, 3], thereby indicating no adverse effects of reduced initial glucose.



## Cultivation Methods

Batch cultivations with two different initial glucose concentrations, 20 mmol l<sup>-1</sup> (n = 3) and 40 mmol l<sup>-1</sup> (n = 3), were performed using CHO cells. At the beginning of the cultivation, only headspace aeration with air was performed until the dissolved oxygen dropped to 40 %. Then, pure oxygen was supplied through a ring sparger in conjunction with headspace aeration to maintain the dissolved oxygen level at 40 ± 1 %. The  $q_{OUR}$  was calculated as explained above.



## Conclusion

The impact of the initial glucose concentration on cell metabolic activity was identified using BlueSens off-gas analyser.

- ✓ Simplified off-gas analytics supports knowledge-based process development
- ✓ Higher  $q_{OUR,max}$  observed for lower initial glucose concentrations
- ✓ Cells take up lactate more actively for lower initial glucose concentrations

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## References

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