An Example of Computational Fluid Dynamics in Biotechnology: Modeling and Simulation of Bacterial Metabolism in Calorimetric Ampule Technique

D. Bethke[1], P. Farber[1], J. Lankowski[1], T. Maskow[2], P. Ueberholz[1]

[1] IMH - Institute of Modelling and High-Performance Computing, Hochschule Niederrhein, Krefeld, Germany

[2] Helmholtz Centre for Environmental Research - UFZ, Leipzig, Germany

Agenda

- The IMH in a Few Words
- About Biotechnology
- Calorimetric Ampule Technique
    - Starting Point
    - Target of the Project
    - Mathematical Model
    - Results of the Simulations
    - Conclusion

The IMH in a Few Words

- People:
  - 4 professors (Parallel Systems, Robust Design Optimization, Mathematics, Optimization, FEM, FSI and CFD)
- Computer:
  - Several compute cluster, the newest has 512 cores and 2 TB RAM
  - Workstation up to 128 GB RAM and 16 cores / high-end GPU's
- Self established mathematical models and program extensions to Fluent

Agenda

- The IMH in a Few Words
- **About Biotechnology**
- Calorimetric Ampule Technique
  - Starting Point
  - Target of the Project
  - Mathematical Model
  - Results of the Simulations
  - Conclusion

About Biotechnology - 1

- **Definition of biotechnology:**
- **"Biotechnology" means any technological application that uses biological systems, living organisms, or derivatives thereof, to make or modify products or processes for specific use. (UN Convention on Biological Diversity, Art. 2)**

About Biotechnology - 2

- Industrial biotechnology is one of the key enabling technologies (KET) of the European Union (see „Key Enabling Technologies - European Commission“)
- KETs are a group of six technologies: micro and nanoelectronics, nanotechnology, industrial biotechnology, advanced materials, photonics, and advanced manufacturing technologies
- KETs have applications in multiple industries and help tackle societal challenges
- Countries and regions that fully exploit KETs will be at the forefront of creating advanced and sustainable economies

About Biotechnology - 3

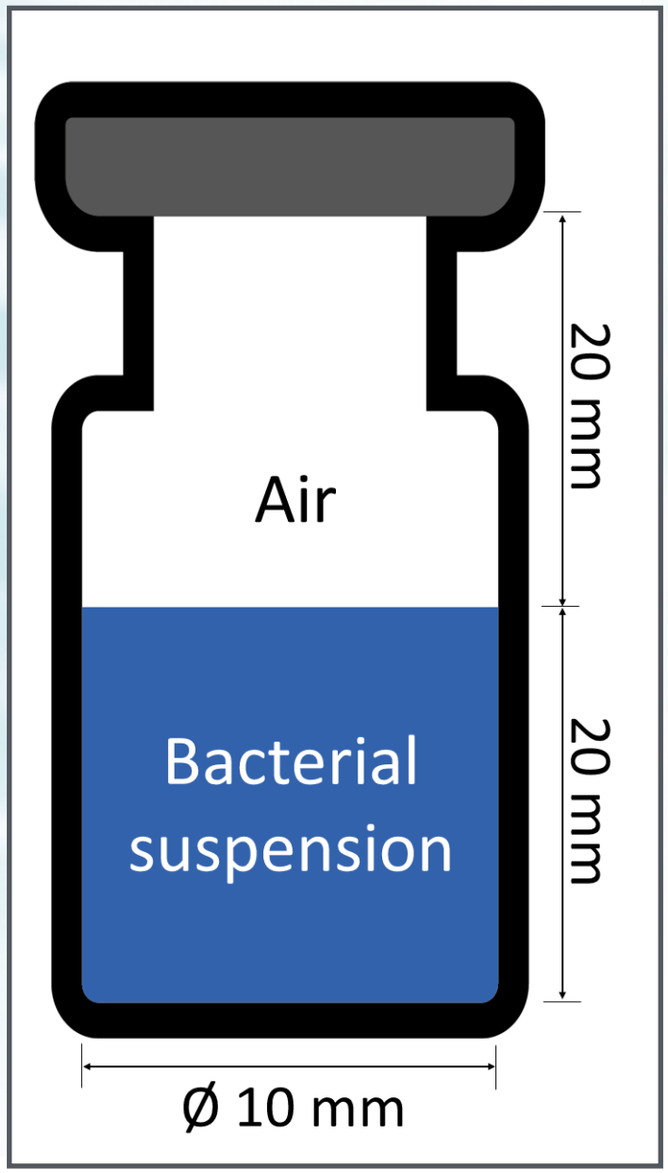
- The biotechnological processes modeled and simulated in our institute:
- Textile Carbon Fibre Electrodes for Microbial Fuel Cells
- Liver-on-a-chip
- Bio-solar cell
- Bio-reactor for the Conversion of C1 Gases
- Calorimetric Ampule Technique

Agenda

- The IMH in a Few Words
- About Biotechnology
- Calorimetric Ampule Technique
  - Starting Point
  - Target of the Project
  - Mathematical Model
  - Results of the Simulations
  - Conclusion

Starting Point - 1

- For calorimetric ampule technique the reaction vessels are filled with cell suspensions and
- the heat exchange with a defined environment is measured under nearly constant temperatures



Starting Point - 2

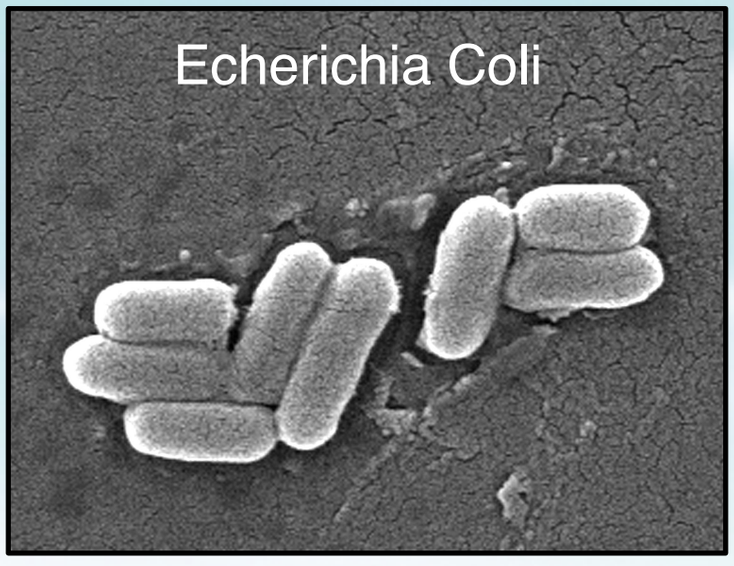
- The calorimetric ampule techniques have found several applications:
- Process optimization in the industrial biotechnology
- Sterility testing in the pharmaceutic and food industry
- Sterility testing and identification of contaminations in medical science
- Death kinetics of cells (e.g. for test of cancer medicine)

Agenda

- The IMH in a Few Words
- About Biotechnology
- Calorimetric Ampule Technique
  - Starting Point
  - Target of the Project
  - Mathematical Model
  - Results of the Simulations
  - Conclusion

Target of the Project

- **Modeling and simulation of the fluid flow, heat transport, the concentration of O_2 and glucose as well as the increase of the number of bacteria cells (*Echerichia coli*)**
- **The CFD simulations are aiming at a better correlation between the calorimetric experiment and the metabolism under investigation than former evaluations**



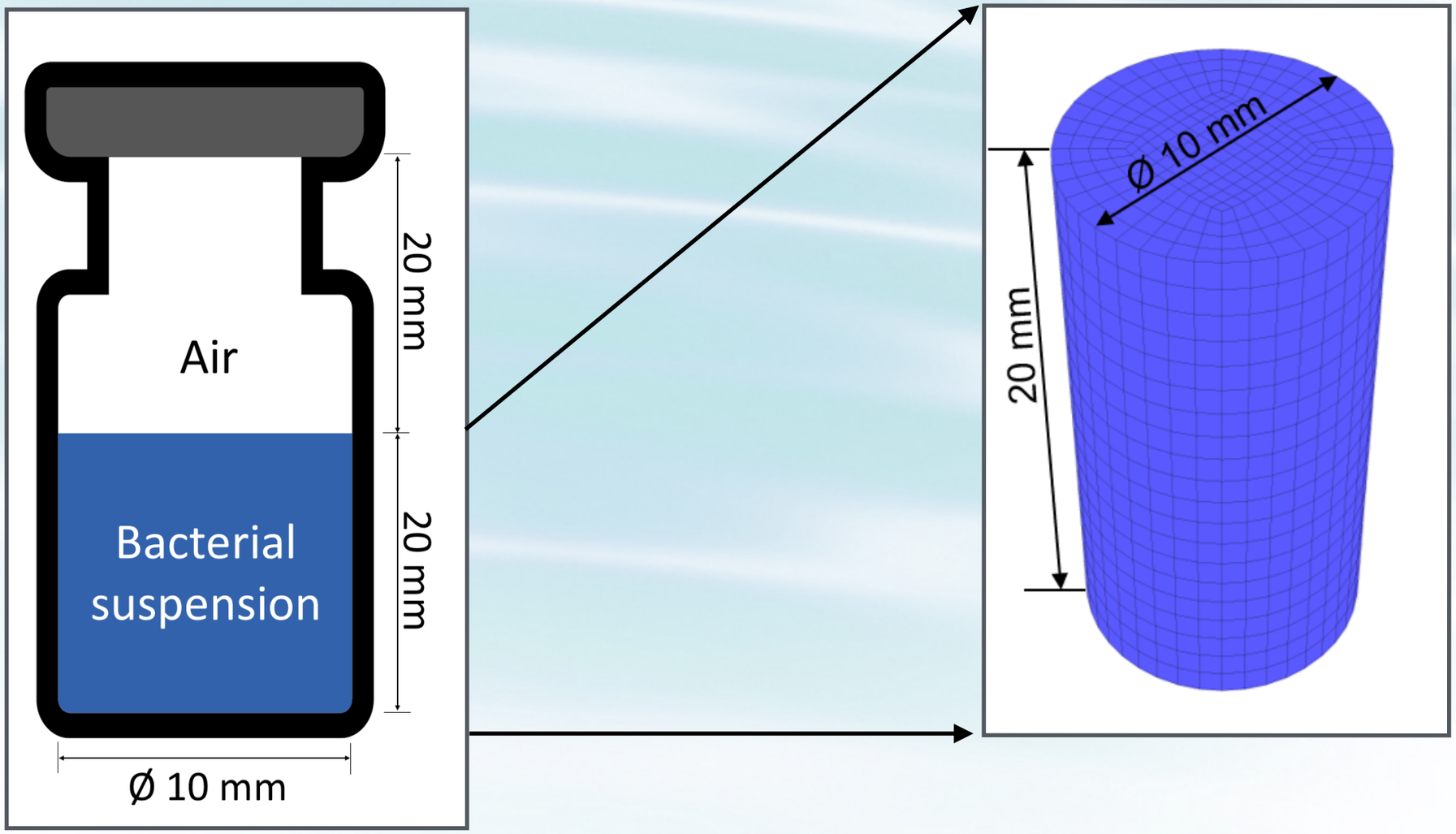
https://pixnio.com/science/microscopy-images/escherichia-coli/this-scanning-electron-micrograph-sem-depicted-a-number-of-gram-negative-escherichia-coli-bacteria 1.3.2018

Agenda

- The IMH in a Few Words
- About Biotechnology
- Calorimetric Ampule Technique
  - Starting Point
  - Target of the Project
  - Mathematical Model
  - Results of the Simulations
  - Conclusion

Mathematical Model - 1

- Until now only the bacterial suspension is modeled and the air is neglected
- Numerical mesh and times step size: Result of a convergence study



Mathematical Model - 2

- **The mathematical model includes so far the fluid (water), modeled in Eulerian description**

Total mass balance equation

Total momentum balance equation

Species O_2 mass balance equation

Species glucose mass balance equation

Energy equation

and the bacteria cells, modeled as spheres of diameter of 1.4 micrometer in Lagrangian description

Sum of forces on the particle = mass of particle * acceleration of particles

Equation for the increase of number of bacteria cells

Energy equation

Mathematical Model - 3

- **The mathematical model includes so far the fluid (water), modeled in Eulerian description**

ρ Density of the phase water with O_2 and glucose

Index O_2: Species O_2

Index g: Species glucose

Index w: Species water

Mathematical Model - 4

- **The mathematical model includes so far the fluid (water), modeled in Eulerian description**

$$\frac{\partial \rho}{\partial t}+\nabla\cdot\left(\rho\vec{v}\right)=0 \qquad \rho=\rho_w$$

$$\frac{\partial\left(\rho Y_{O_2}\right)}{\partial t}+\nabla\cdot\left(\rho\vec{v}Y_{O_2}\right)=\nabla\cdot\vec{J}_{O_2}+S_{m,O_2}$$

$$\frac{\partial \rho\vec{v}}{\partial t}+\nabla\cdot\left(\rho\vec{v}\otimes\vec{v}\right)=-\nabla p+\nabla\cdot\vec{\vec{T}}+\left(\rho-\rho_{op}\right)g+\vec{F}_b$$

$$\frac{\partial\left(\rho Y_g\right)}{\partial t}+\nabla\cdot\left(\rho\vec{v}Y_g\right)=\nabla\cdot\vec{J}_g+S_{m,g}$$

$$\frac{\partial}{\partial t}\left[\rho E\right]+\nabla\left[\vec{v}\left(\rho E+p\right)\right]=\nabla\cdot\left(k\nabla T\right)-\nabla\left(h_{O_2}\vec{J}_{O_2}+h_g\vec{J}_g\right)+S_{h,b}$$

$$\vec{F}_b=-\frac{1}{V_c}\sum\left(\frac{\vec{v}-\vec{v}_b}{\tau_r}\right)m_b$$

Mathematical Model - 5

The bacteria cells, modeled as spheres of diameter of 1.4 micrometer in Lagrangian description

$$\frac{\partial \vec{v}_b}{\partial t} = \frac{\vec{v} - \vec{v}_b}{\tau_r} + \frac{\vec{g}\left(\rho_b - \rho_2\right)}{\rho_b} \qquad \tau_r = \frac{\rho_b d_b^2}{18\mu_2}\frac{24}{C_d \mathrm{Re}_b} \qquad C_d = \frac{24.0}{\mathrm{Re}_b} \qquad (\mathrm{Re_b} < 0.1) \qquad \mathrm{Re}_b = \frac{\rho_2 d_b \left|\vec{v}_b - \vec{v}_2\right|}{\mu_2}$$

$$\Delta n_b V_c \cong \frac{R_{Max}^{Aerob} Y_{O_2} + R_{Max}^{Anaerob} Y_{O_2,1/2}}{Y_{O_2,1/2} + Y_{O_2}} \cdot \frac{Y_g}{Y_{g,1/2} + Y_g} \cdot n_b \cdot \Delta t \cdot V_c$$

$$m_b c_{p,b} \frac{dT_b}{dt} = A_b h\left(T - T_b\right) + S_{h,O_2} \qquad h = \frac{k_2}{d_b} Nu_b = \frac{k_2}{d_b}\left(2.0 + 0.6\mathrm{Re}_b^{\frac{1}{2}} \mathrm{Pr}^{\frac{1}{3}}\right)$$

Mathematical Model - 6

- **Sinks and sources - fluid equations**

  Mass sink for O_2 in the O_2 species balance equation

$$S_{m,O_2} \cong -C_{m,O_2} R_{Max} n_b \frac{m_p}{M_{CH_{1.70}O_{0.42}N_{0.25}}} \frac{Y_{O_2}}{Y_{O_2,1/2}+Y_{O_2}} M_{O_2}$$

  Mass sink for glucose in the glucose species equation

$$S_{m,g} \cong -\frac{Y_{O_2,1/2}+Y_{O_2}}{C_{m,g}^{Aerob} Y_{O_2} + C_{m,g}^{Anaerob} Y_{O_2,1/2}} \cdot \frac{R_{Max}^{Aerob} Y_{O_2} + R_{Max}^{Anaerob} Y_{O_2,1/2}}{Y_{O_2,1/2}+Y_{O_2}} \cdot \frac{Y_g}{Y_{g,1/2}+Y_g} \cdot n_b \frac{m_p}{M_{CH_{1.70}O_{0.42}N_{0.25}}} \cdot M_{C_6H_{12}O_6}$$

Thermal energy source in the fluid energy equation

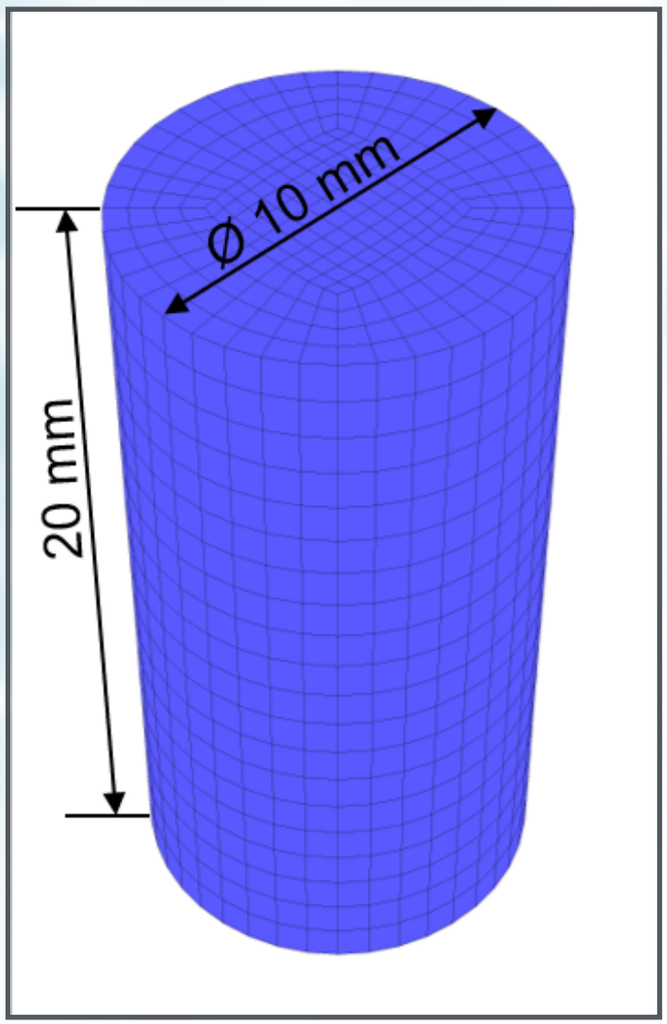
$$S_{h,b} = A_b h\left(T - T_b\right)$$

Thermal energy source in the particle energy equation

$$S_{h,O_2} = -\frac{C_{h,O_2}}{M_{O_2} n_b} S_{m,O_2}$$

Mathematical Model - 7

- All walls are modeled as no slip wall
- For the species balance equation of glucose all walls are impermeable
- For the balance equation of O_2 all walls except the top wall are impermeable
- At the top wall the equilibrium mass fraction of O_2 with air is set
- The boundary condition for the particles at the walls is reflect

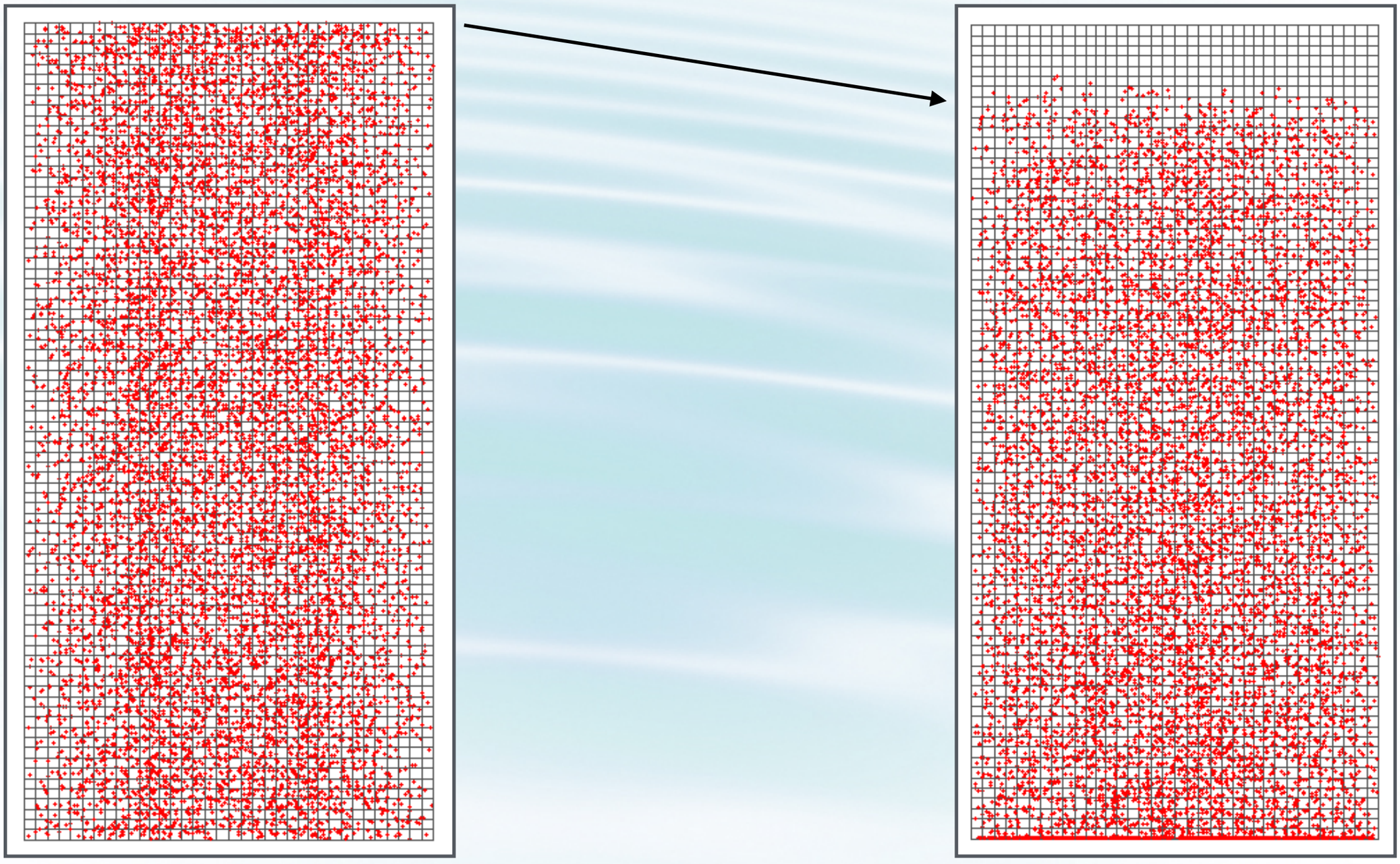


Agenda

- The IMH in a Few Words
- About Biotechnology
- Calorimetric Ampule Technique
  - Starting Point
  - Target of the Project
  - Mathematical Model
  - Results of the Simulations
  - Conclusion

Results - Sedimentation of the Bacteria Cells

- **Due to the higher density of the bacteria cells they settle approx. 2 mm over 5 hours**

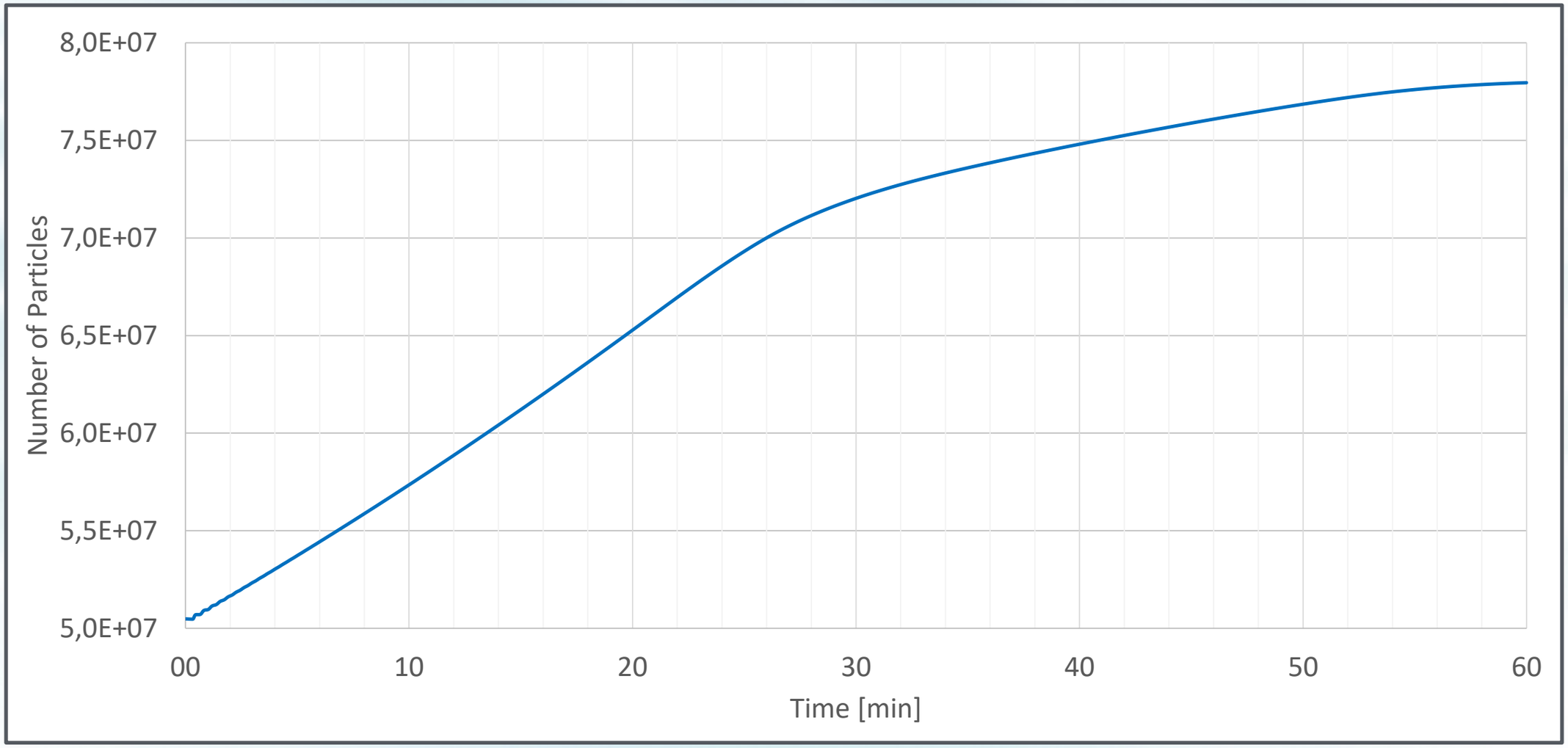


t = 1 s

t = 18000 s

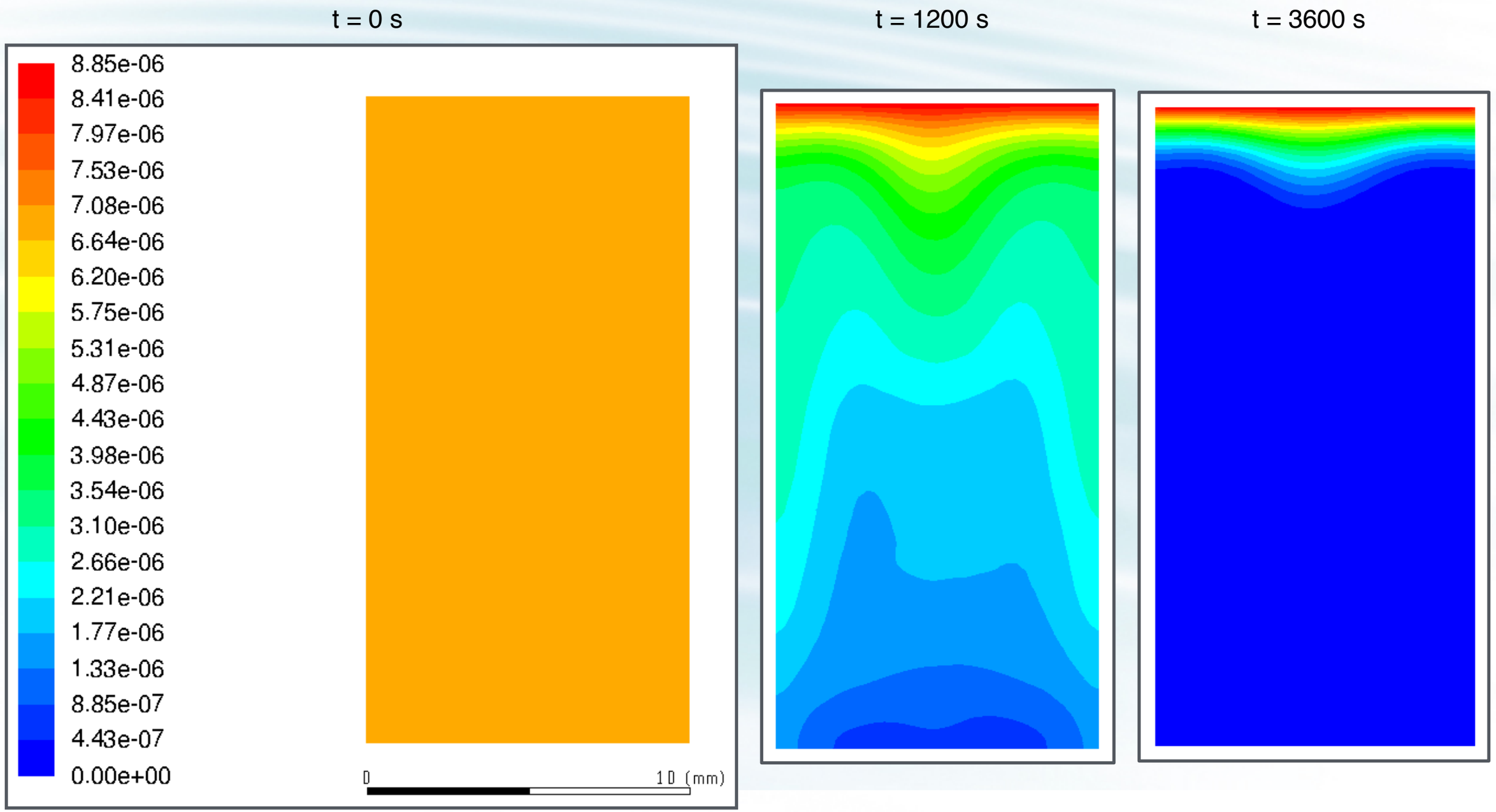
Results - Increase of Number of Bacteria Cells

- **As a function of consumption of O_2 and glucose the number of bacteria cells growths**



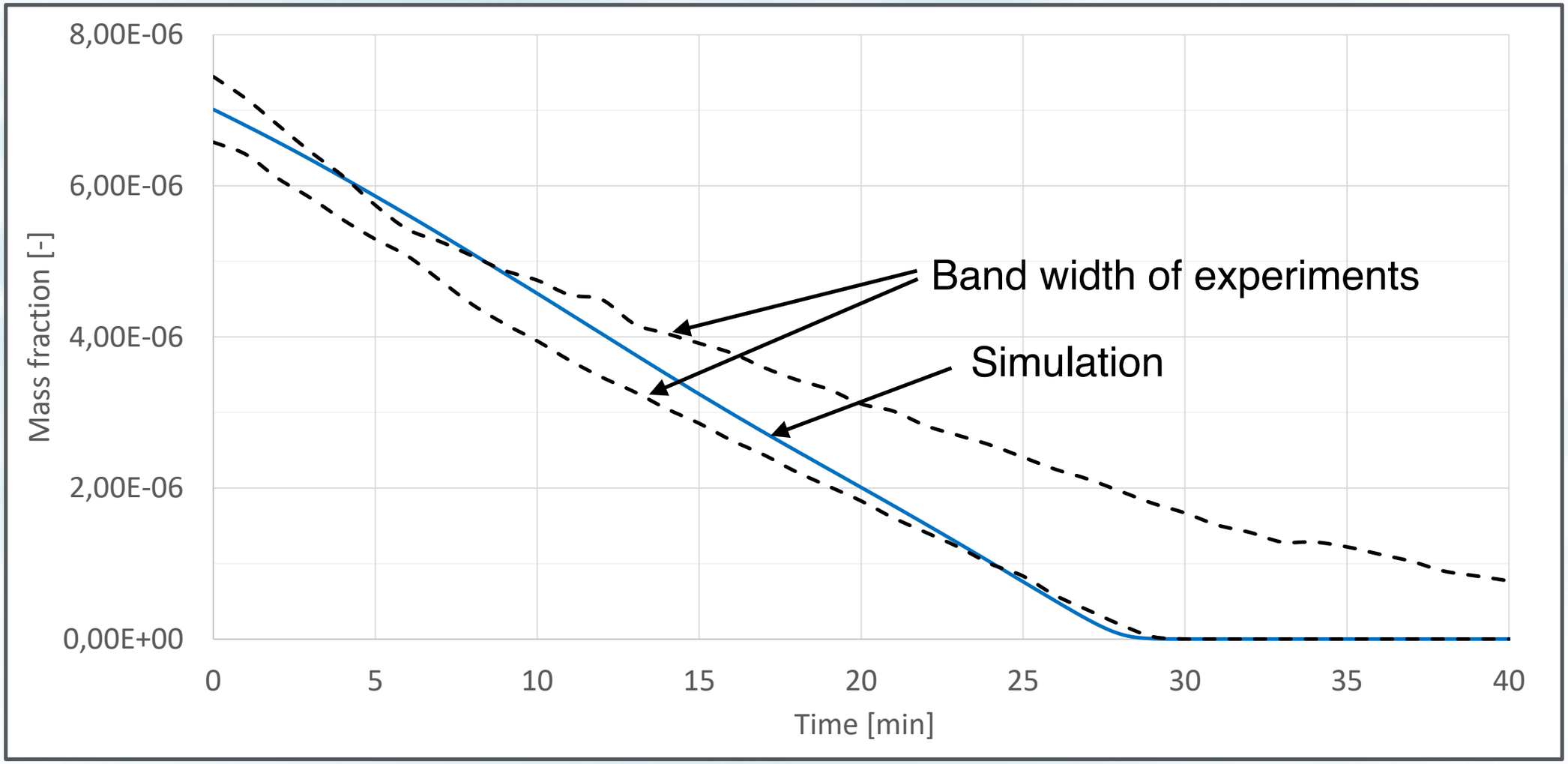
Results - Depletion of O_2 - 1

- **The mass fraction of O_2 is decreasing because of the consumption by the bacteria cells**



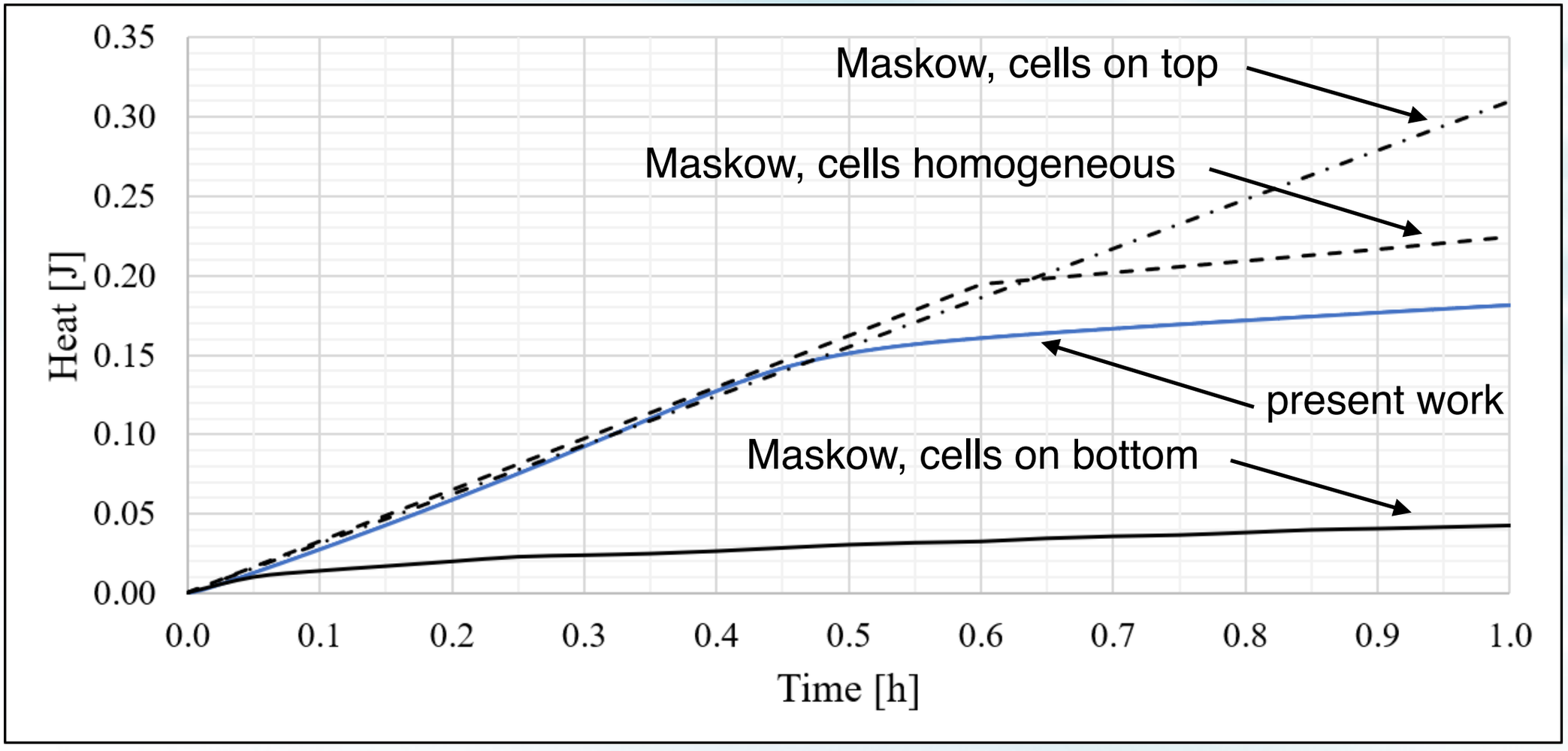
Results - Depletion of O_2 - 2

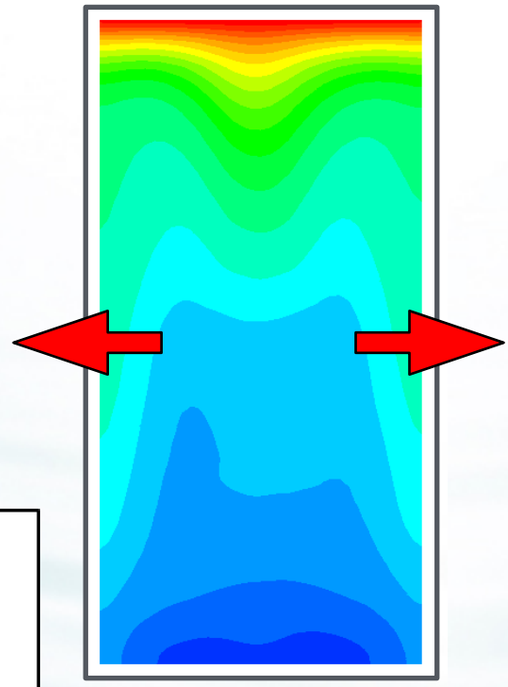
- **Measurements in the center of the ampule liquid spread and the simulation matches them**



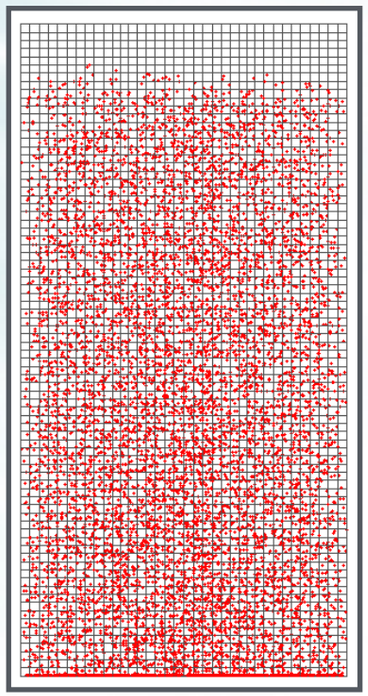
Results - Generation of Thermal Energy by Bacteria

- **Heat exchanged with the environment**
- **Comparison with bacteria modeled as a dissolved species (Maskow et al., 2014)**





Mass fraction of O_2 after 1200 s



Heat production is a function of location of cells

Agenda

- The IMH in a Few Words
- About Biotechnology
- Calorimetric Ampule Technique
  - Starting Point
  - Target of the Project
  - Mathematical Model
  - Results of the Simulations
  - Conclusion

Conclusion- 1

- **Conclusions for calorimetry users:**
  - Take care with the thermokinetic interpretations, because the heat signal is corrupted by oxygen limitations and biomass sedimentation
  - Try to reduce the parasitic effects already in the experimental design (by growth on agar, low biomass concentrations, adjustment of the density of the medium etc.)

Conclusion- 2

- **Conclusions for calorimetry manufacturers:**
  - Develop multichannel calorimeters where the bacterial suspension can be homogeneously mixed (shaken or stirred)
  - Develop solutions for oxygenation without disturbing evaporation effects

Conclusion- 3

- General conclusion:
- Biotechnological processes are in the reach of being modeled and simulated by CFD
- This generates new insights into the processes
- Possible with tools like Fluent, when biotechnology specific models and software routines are added

References

Maskow T, Morais FM, Rosa LFM, Qian, Harnisch F (2014) Insufficient oxygen diffusion leads to distortions of microbial growth parameters assessed by isothermal microcalorimetry, RSC Adv. 4:32730–32737.

Key Enabling Technologies - European Commission, http://ec.europa.eu/growth/industry/policy/key-enabling-technologies_en (access on 22/8/2018).

UN Convention on Biological Diversity, Art. 2, https://www.cbd.int/convention/articles/default.shtml?a=cbd-02 (access on 22/8/2018).

Hochschule Niederrhein
University of Applied Sciences

IMH
Institut für Modellbildung
und Hochleistungsrechnen
Institute of Modelling
and High-Performance Computing

http://www.hs-niederrhein.de/forschung/imh/

peter.farber@hs-niederrhein.de