



EVOLUTION OF TECHNOLOGY

Customised technology for pharmaceutical and biotechnological plants

Birgit Pittermann, Dublin, Oct 20th 2016

- Short overview about ZETA
- ZETA production units for biotechnological processing
- ZETA Freezing process development
- ZETA mixing performance

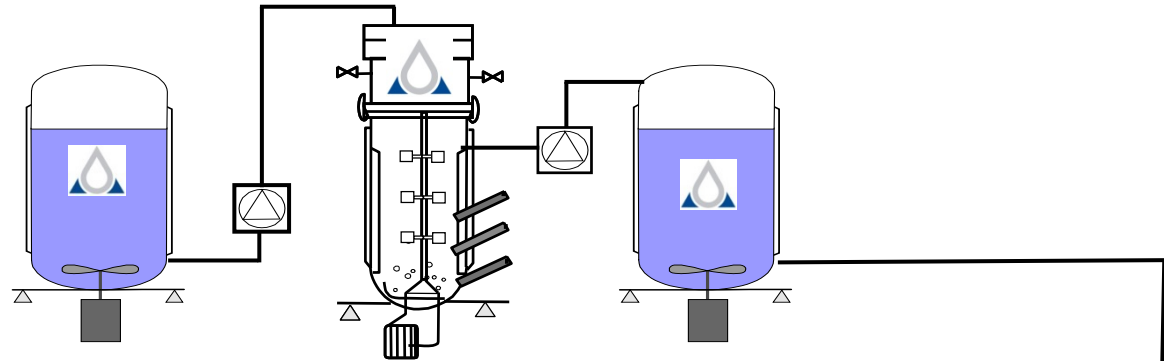


- Short overview about ZETA
- **ZETA production units for biotechnological processing**
- ZETA Freezing process development
- ZETA for mixing technologies



**Upstream
Process
(USP)**

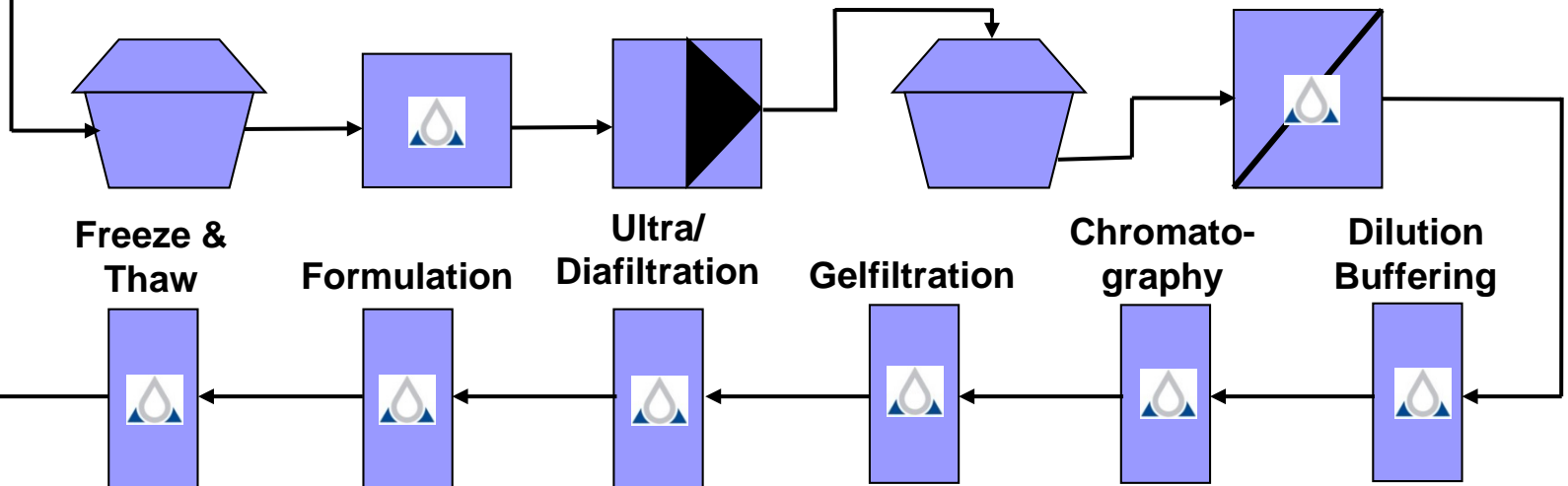
Substrate Tank Bioreactor Harvest Tank



Fermentation

**Downstream
Process
(DSP)**

Centrifugation Cooling Homogenisation Centrifugation Filtration



Project Example: Freeze Controller

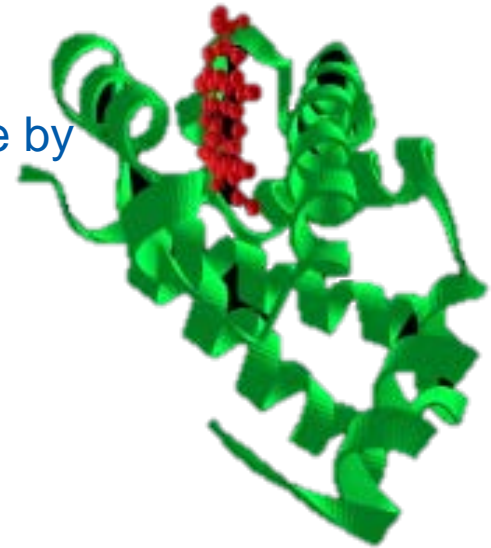


- Short overview about ZETA
- ZETA production units for biotechnological processing
- **ZETA Freezing process development**
- ZETA mixing technologies



Initial Situation

- Protein APIs are a growing market for the pharmaceutical industry
- In 2018 80 % of new drugs will be large molecules
- Protein storage and transport at bulk level is mostly done by freezing
- Freezing can lead to protein damage, which may lead to
 - Negative immune reaction
 - Loss of activity
- Freezing process and changes to protein solution is rarely defined
- Process control influences protein quality



Cold Denaturation

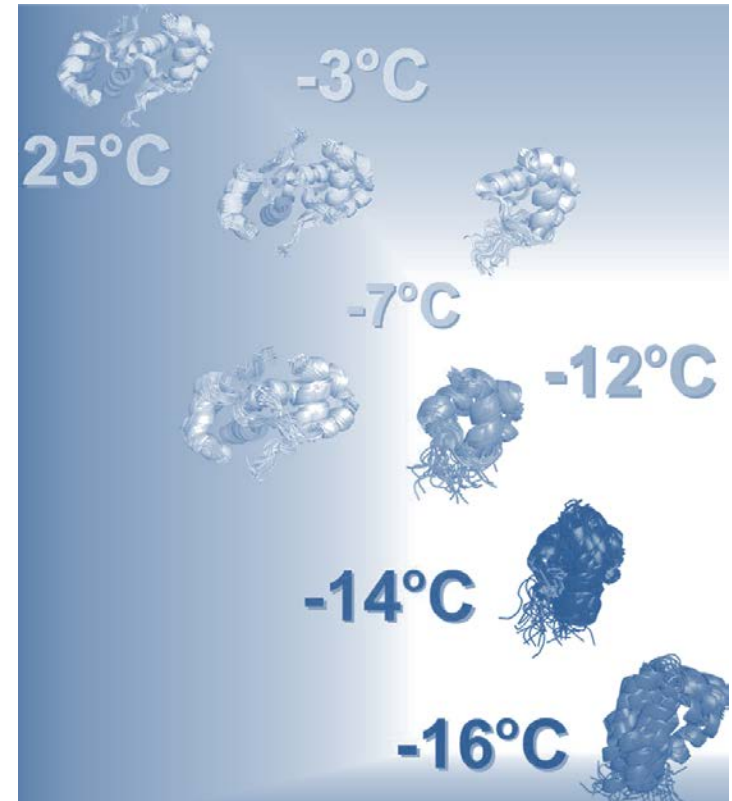
- Reversible

Cryoconcentration

- Proteins, co-solutes, buffer salts
- Precipitation of proteins, crystallization, pH-shift
- Phase separation
- Change of solubility of proteins and salts

Surface Stresses

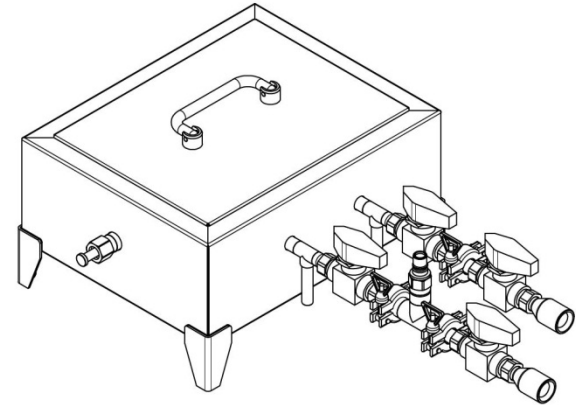
- Ice surface, insoluble additives

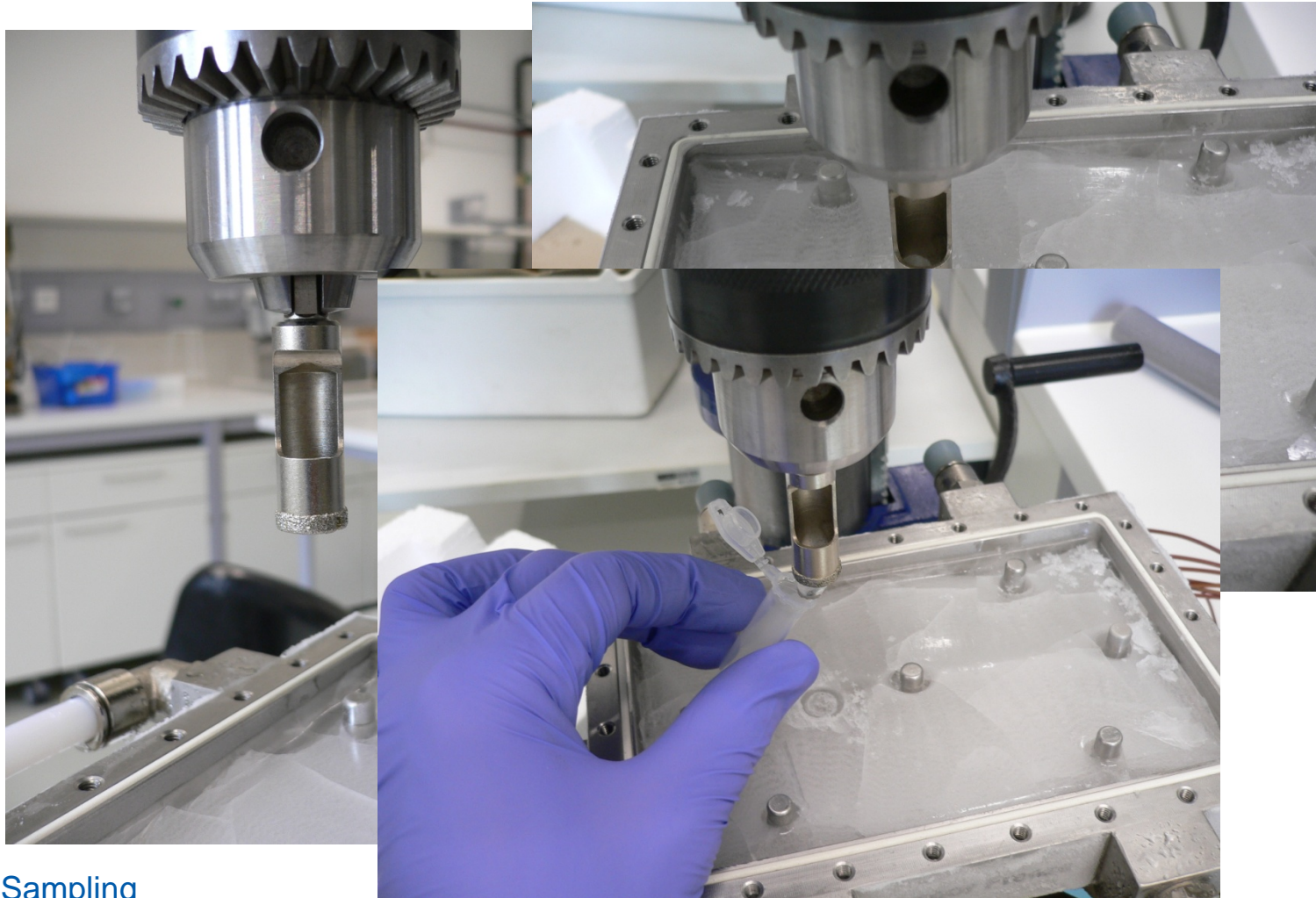


Few general rules => high variability in proteins and co-solvents

Lab-Scale Freeze Container

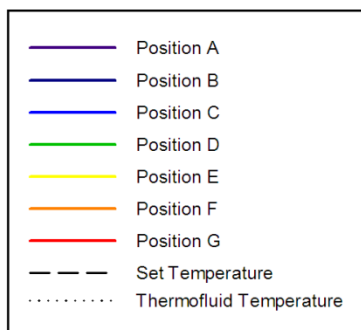
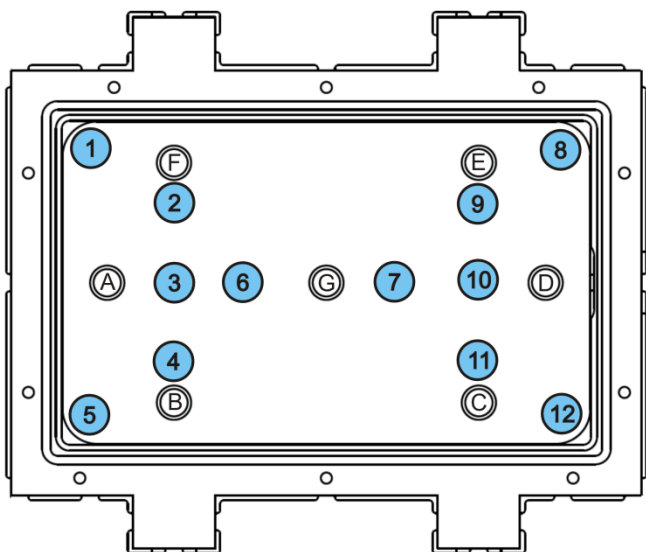
- Working volume: 200 ml
- Overall sampling
- 7 temperature sensors for online monitoring
- Removal of ice-block possible



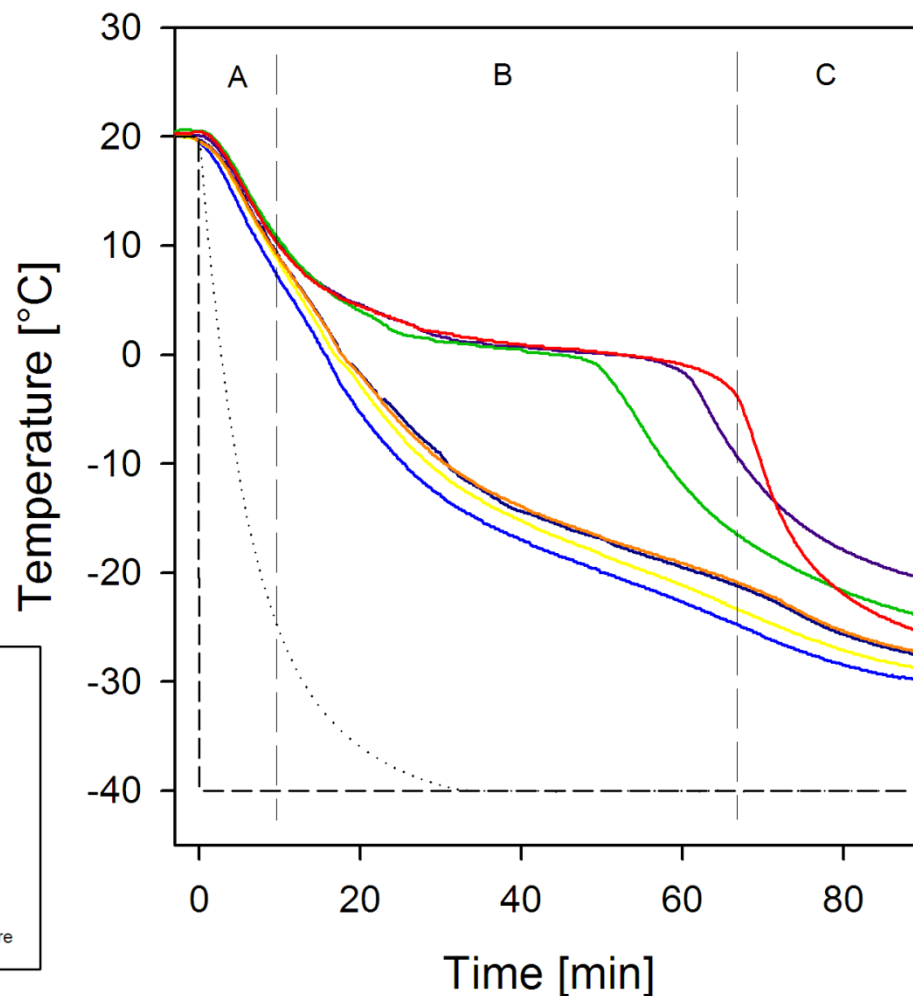


Sampling

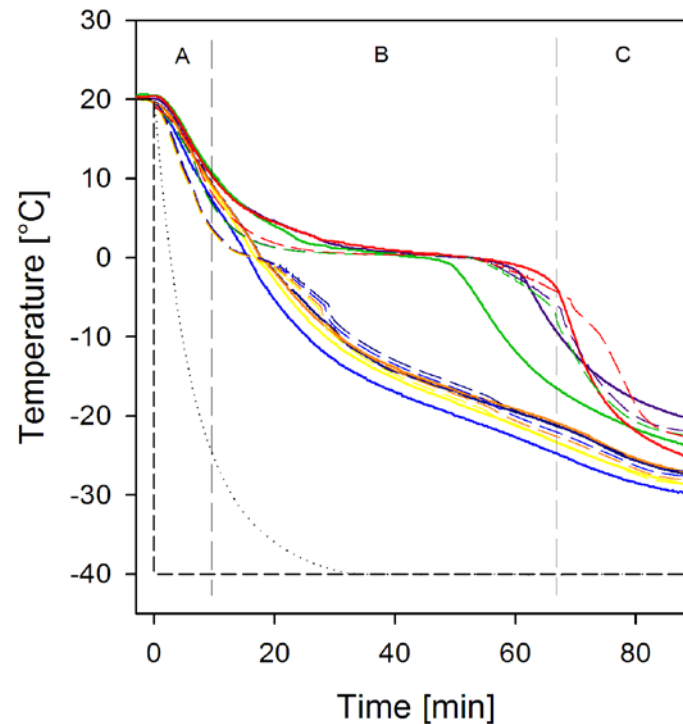
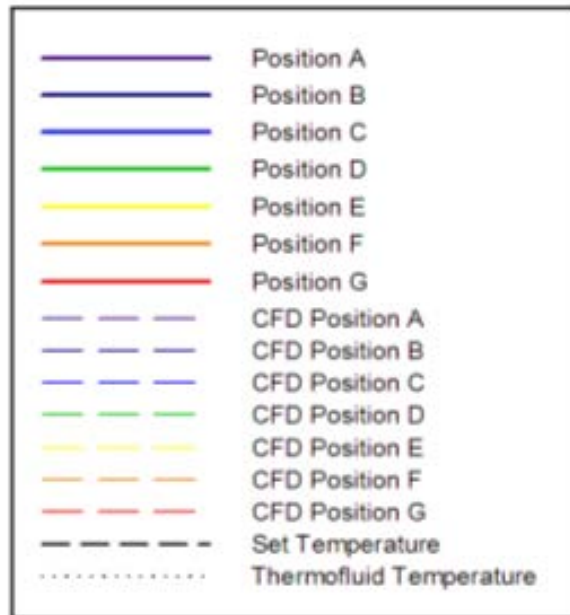
The Freezing Process



A – Initial cooling phase
B – Freezing phase
C – Equilibration phase

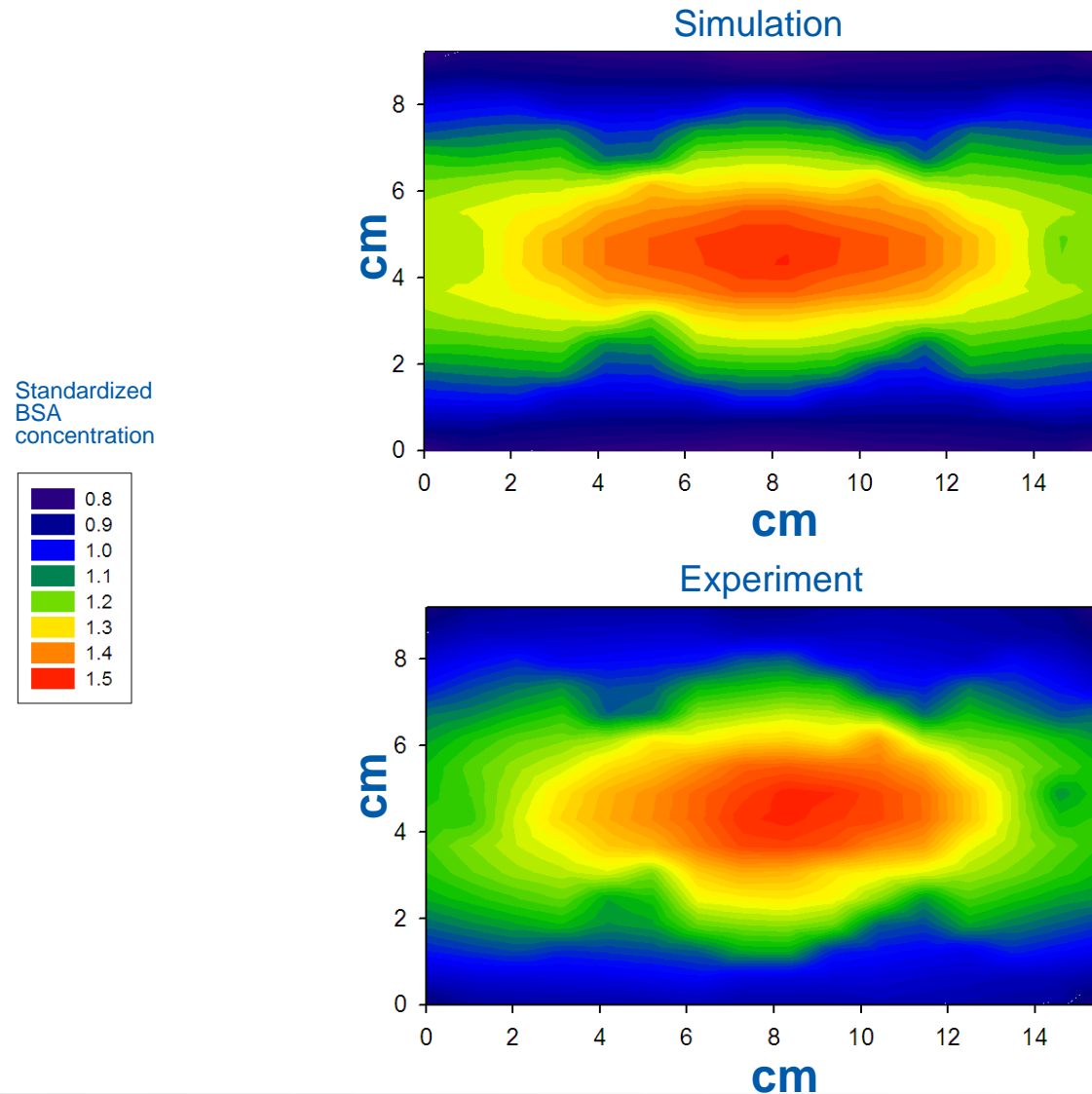


- Temperature and ice front propagation
BSA, 100 µg/ml, phosphate buffer
7 temperature probes, 24 samples



CFD simulations of the freezing process

Distribution of BSA concentration



700 ml scale-down model

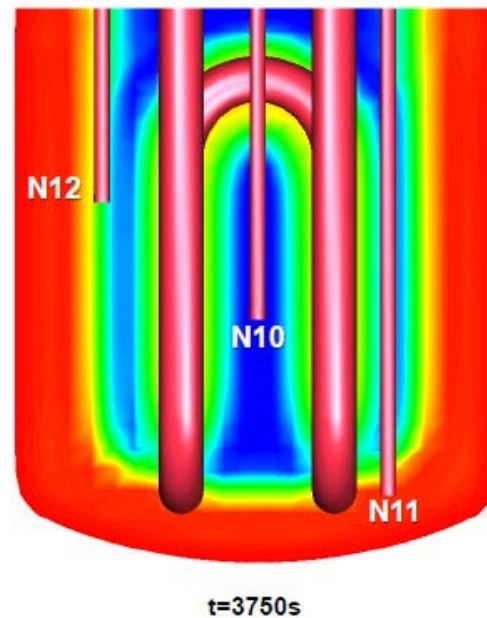
- Stainless steel
- External cooling
- Multiple temperature sensors
- pH monitoring
- Recirculation of the solution possible during F/T
- Sampling and investigation of whole block of ice possible
- Mobile construction

Design of Pilot Scale Freezer is based on lab scale experiments, CFD simulation and experience of industrial conditions

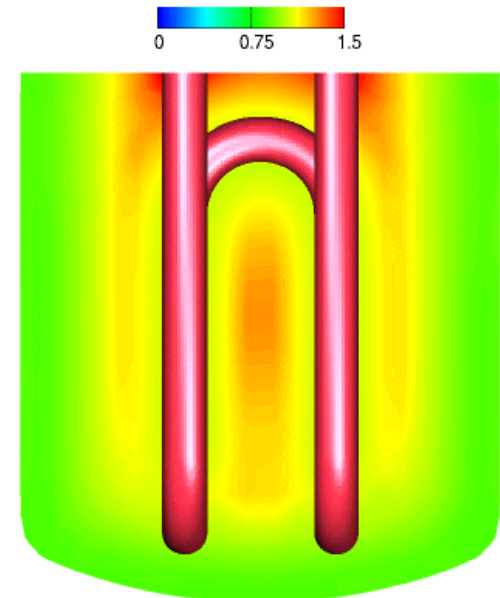


700 ml scale-down model

- CFD Simulation
 - Temperature profile
 - Concentration profile
- Experimental Verification



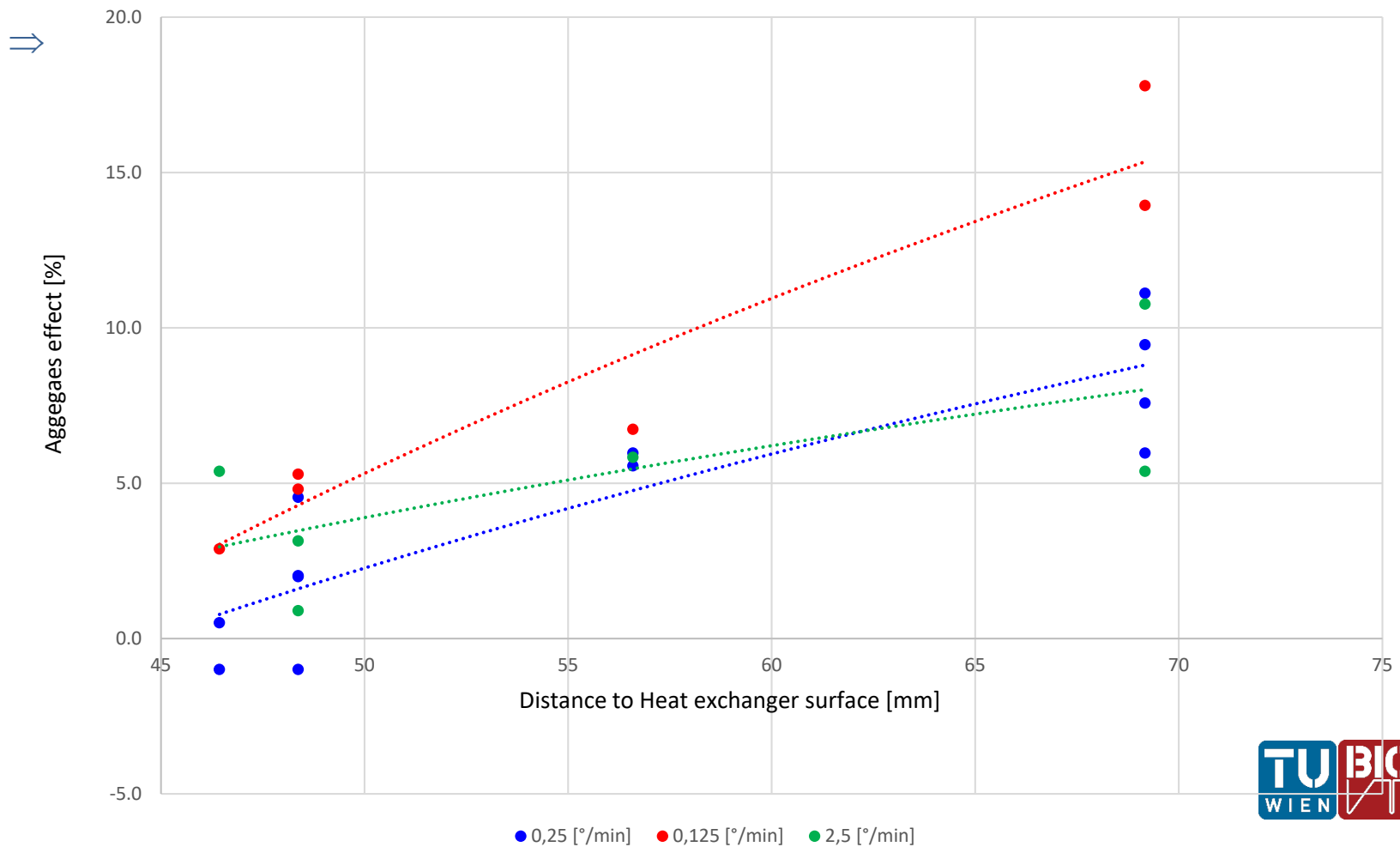
Cryo concentration



Contour plot: cryo concentration factor [-]

Freezing mAb - Results Aggregates

IgG Protein A Aggregates - 280nm



Laboratory freezing equipment 0,2 L



R&D

- Lab-scale
- Product characterization
- Controlled freezing
- CFD simulations

Scale-up

Pilot Freeze Container 0,7 L



Tech Transfer

- Pilot scale
- CFD simulations
- Process development
- Process monitoring

Scale-up

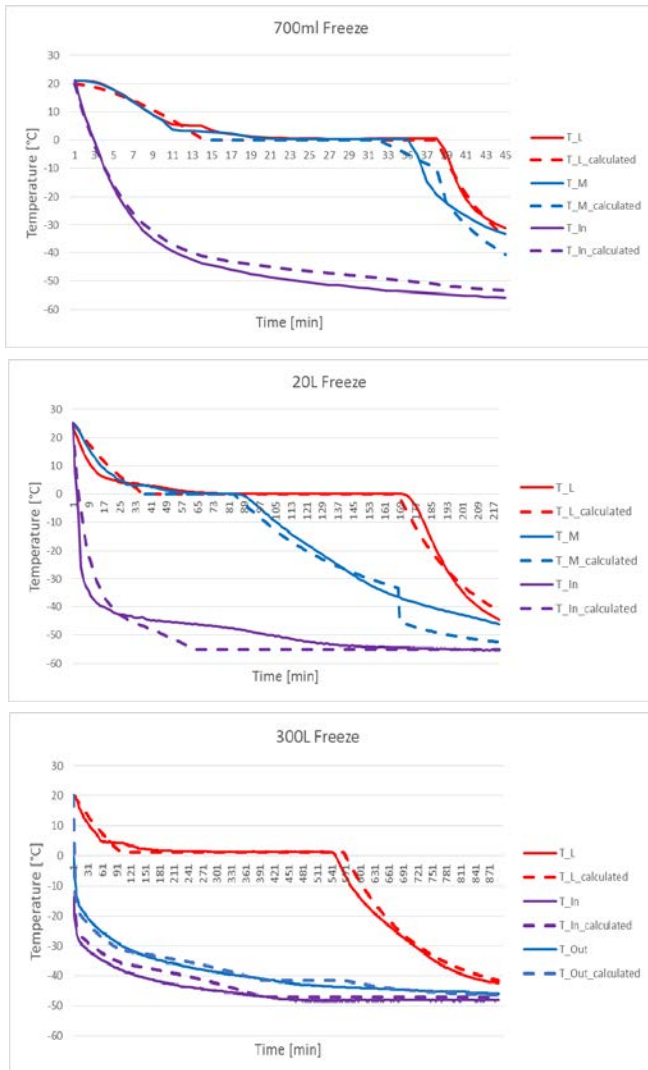
Zeta Freeze Container 300 L



Manufacturing

- Customised scaling of vessel and process
- CFD simulations
- Process monitoring

Freeze Process Simulation



Functions / Process steps

- Freezing
- Thawing
- Blow Down
- Sterilisation

Recipe based configuration of all functions

The following options are currently available:

- Shaker for 300 liter container
- Product pump for circulation and sampling
- Container valve control

Additional options can be integrated over the phase model.





20 ml

Freeze Container



51

- **Enabling a Transparent Freezing Process**
 - Optimisation of freezing process with respect to protein activity
 - Constant product-quality by design met by process knowledge
 - CFD-simulation of temperature, phase and solute distribution
 - Multiple monitoring possibilities
 - Process safety for stainless steel solution
- **Scaling the Process**
 - Scaling possible due to process modeling
 - Customised solutions
 - Production scale unit for high quality products
 - CFD-based container design and probe placement
 - Considering critical process parameters
 - Consistent freezing solutions from R&D- to manufacturing scale

- Short overview about ZETA
- ZETA production units for biotechnological processing
- ZETA Freezing process development
- **ZETA mixing technologies**





ZETA BMR(T)

Bottom mounted

ZETA BMR- F

for Bioreactors



ZETA AMRi

Top mounted -
magnetic coupling internally

ZETA AMRe

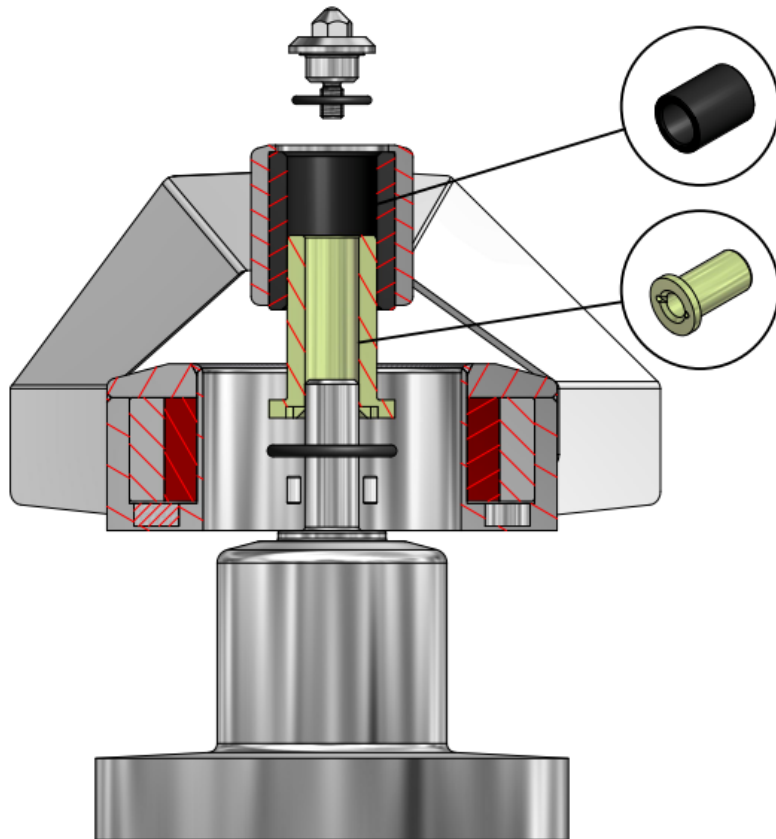
Top mounted -
magnetic coupling externally



At a glance

- **Bottom or top mounted for vessels up to 50,000 L**
- **Frictionless power transmission**
- **CIP/SIP optimized design**
- **Dry run capability**
- **Quick bearing replacement**
- **Shaft extension up to 2 meters**
- **Weld-on or plugged-in solution**
- **Retrofit friendly design**





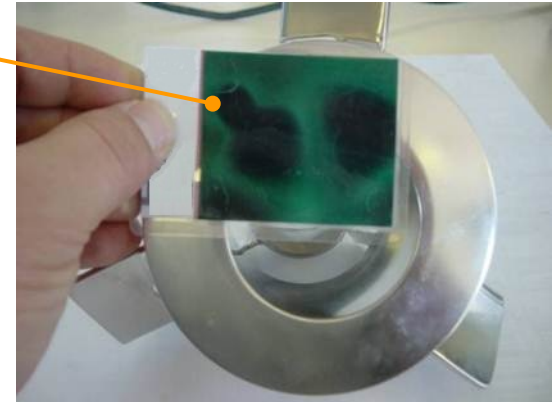
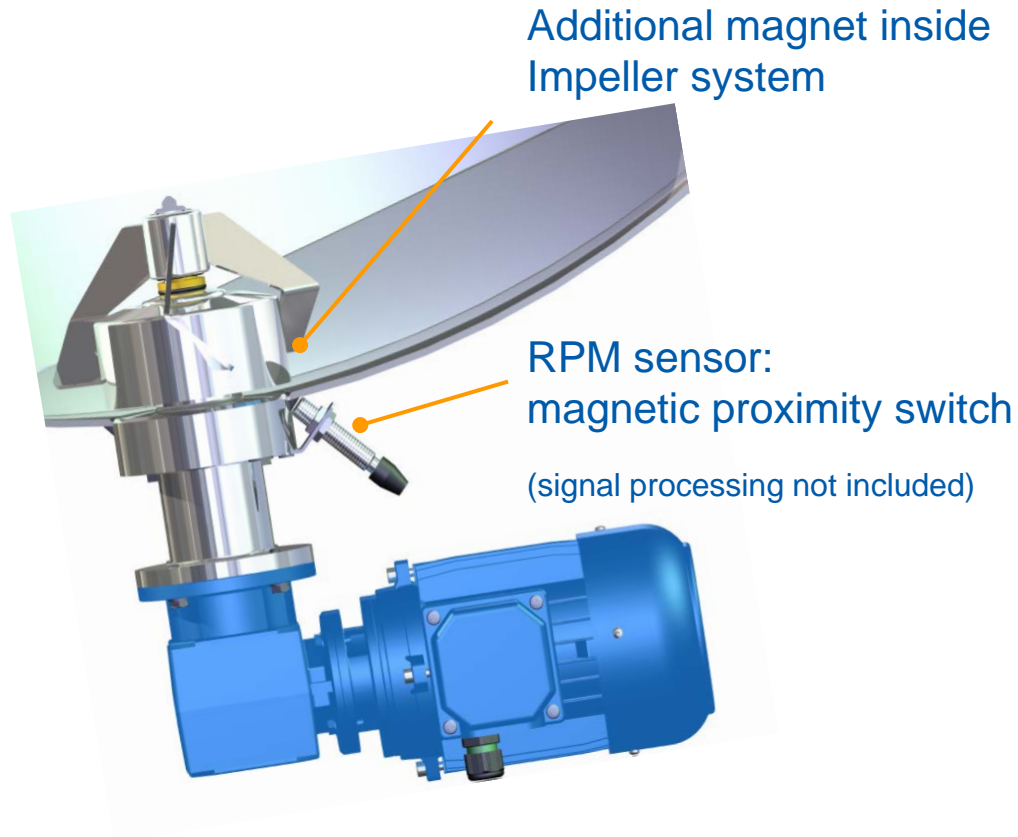
- **Unique ZETA bearing design**
- **SiC rotating bearing featuring lubrication channels to minimize wear**
- **ZrO2 static bearing with high stress tolerance**
- **Anti-twist-protection preventing rotation of the male bearing**
- **Pivot bolt for bearing support showing improved stability being one turned part (weld plate, containment shell, pivot)**

Agitator Types - BMRT

Model no.	Torque max.	Ø Impeller	Mixing capacity in litres (depending on application)	
	Nm	mm	min	max
BMRT 35	0.3	80	2	100
BMRT 50	0.5	80	5	140
BMRT 80 M	0.8	105	50	350
BMRT 125	1.25	130	100	500
BMRT 400	4	165	200	1200
BMRT 800	8	190	700	2500
BMRT 1300	13	250	1000	4500
BMRT 2600	26	300	2000	8000
BMRT 5000	50	350	3500	15000
BMRT 10000	100	400	6000	25000
BMRT 17000	170	450	10000	40000
BMRT 21000	210	500	15000	55000



RPM Measurement



 **Standard for agitators in Ex Zone 0/1**
(for safety reasons)

Substitution of 25 mechanically sealed agitator



Design Daten	Unit	BMR F 13000	BMR F 30000
Vessel diameter	[mm]	2000	2000
filling volume	[L]	12000	15000
Filling height in vessel	[mm]	3986	4293
H/D Ratio		2,0	2,5
speed range	[min ⁻¹]	07 -60	10 – 85
diameter	[mm]	665	665
d/D-ratio	[-]	0,33	0,33
Tip-Speed (at max rpm)	[m/s]	2,02	2,51
Spezif. Leistungseintrag	[kW/m ³]	0,08	0,15

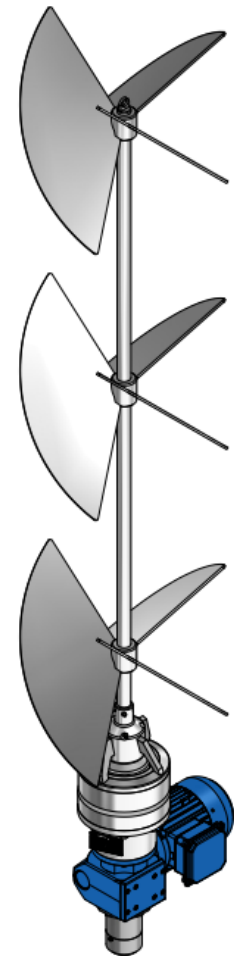
Improvements of:

- ⇒ **sterility**
- ⇒ **less maintenance**

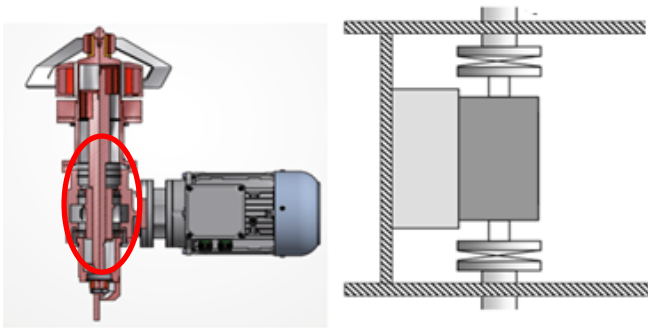
Reduction of:

- ⇒ **maintenance work and time**
- ⇒ **energy consumption**
- ⇒ **production costs**

Agitator	Unit	500 L vessel	2000 L vessel
Mixing task	[-]	cell culture	
Vessel diameter	[mm]	700	1100
Filling height in vessel	[mm]	1351	2186
Filling volume	[L]	500	2000
Agitator rpm	[min ⁻¹]	20- 120	20- 85
Stirrer type	[-]	2 x elephant ear	3x elephant ear
Diameter stirrer	[mm]	305	450
d/D-ratio	[-]	0,44	0,41
max RPM	[min ⁻¹]	120	85
Tip-Speed (at max rpm)	[m/s]	1,92	2,00
Specific power input	[kW/m ³]	0,144	0,146

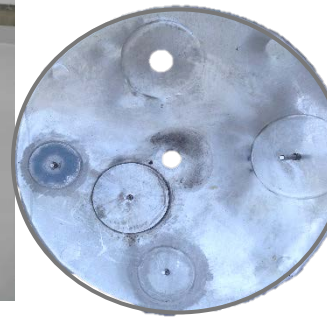


Torque Measurement

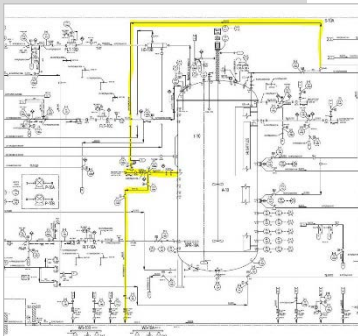


- ✓ Detailed knowledge about Power input to Media
- ✓ Treat cell membranes with highest possible care
- ✓ Best grade of homogeneity in the process
- ✓ Optimum gas bubble dispersion

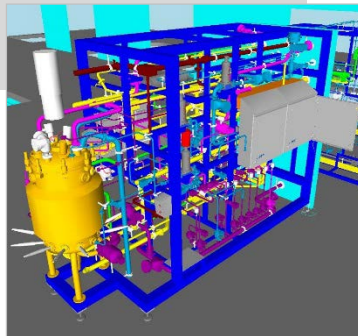
Test Vessel with double jacket



Process



Engineering



Equipment



Service &
Maintenance





Zeta Biopharma – Your Partner for Biotech and Pharma

Thank you for your attention!

Birgit Pittermann, Head of R&D,

Birgit.Pittermann@zeta.com