Investigation of growth processes of bacterial populations by caloric measurements

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Abstract
Bacterial populations growing in substrate including broths always show caloric effects, i. e. exchange of heat with surroundings. This heat can be measured by (isothermal) microcalorimeters and allows to determine the population dynamics of the bacteria. This dynamics is characteristic for the bacteria and allows in principle its rapid identification which is of importance in case of infections for medical diagnostics and therapy.

1. Bacteria

A. van Leeuwenhoek 1676
Microscop

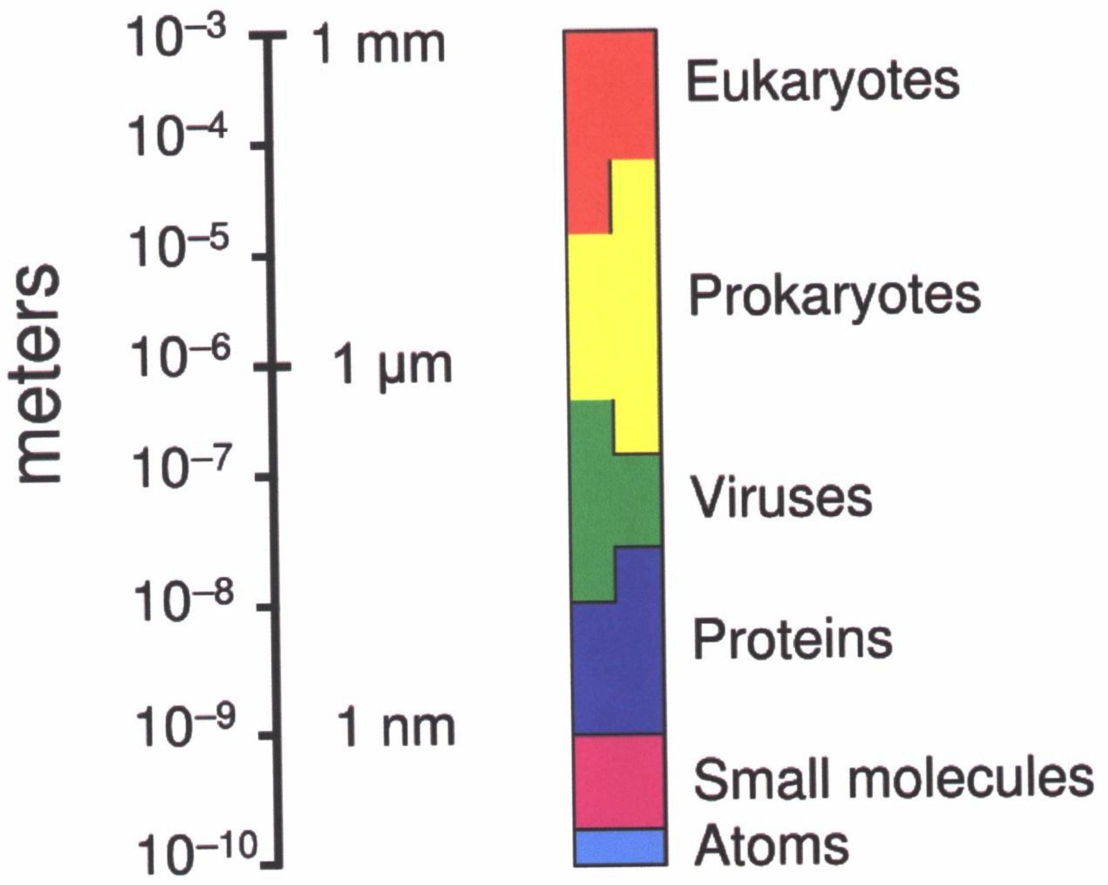
Water
Human spittle

10*9 – 10*12 / liter

Known bacteria 1 – 5 %

Unknown bacteria 99 – 95 %

Mass: 1 – 10 p g, p = 10*-12



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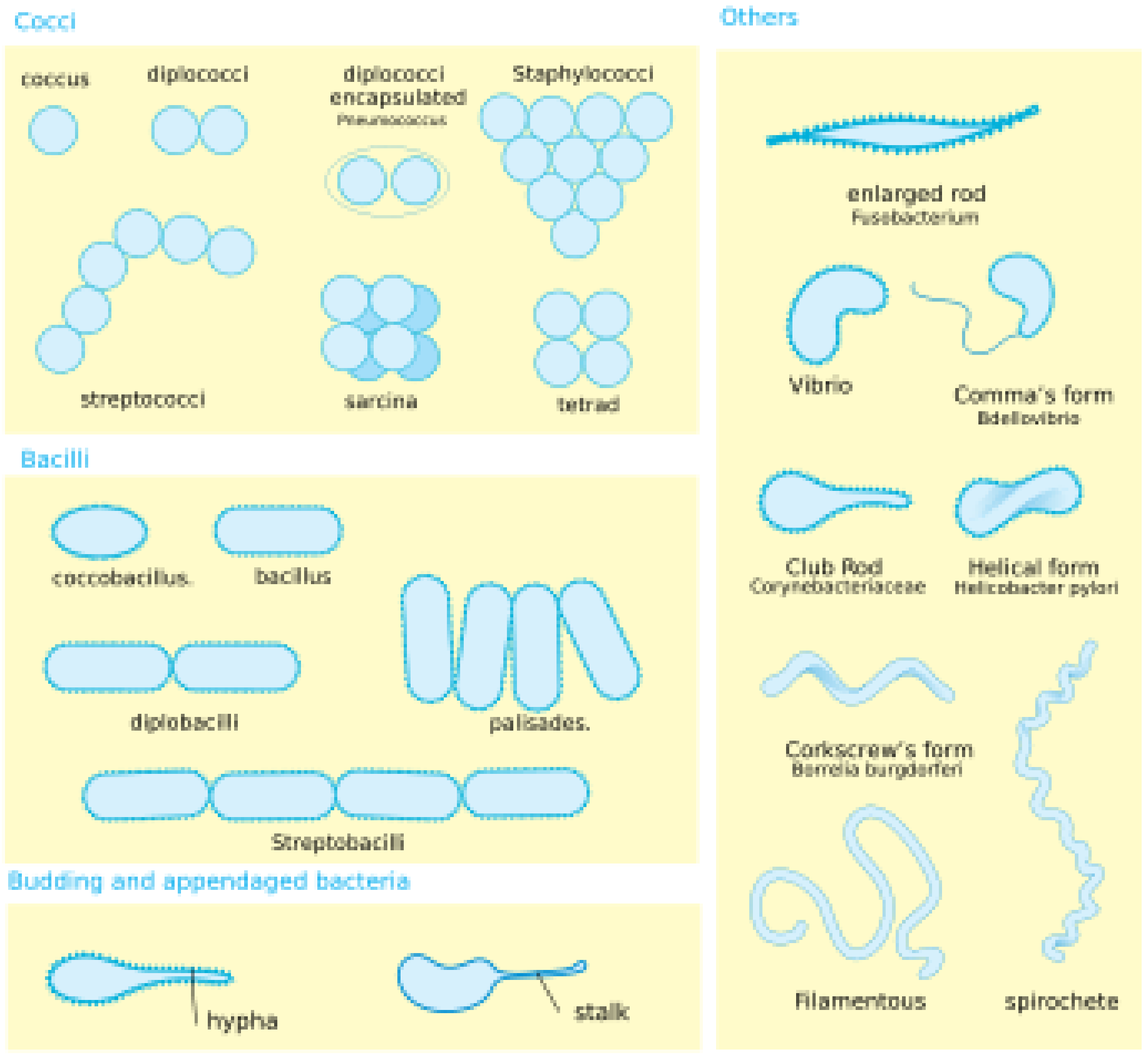
Bacteria, Morphology

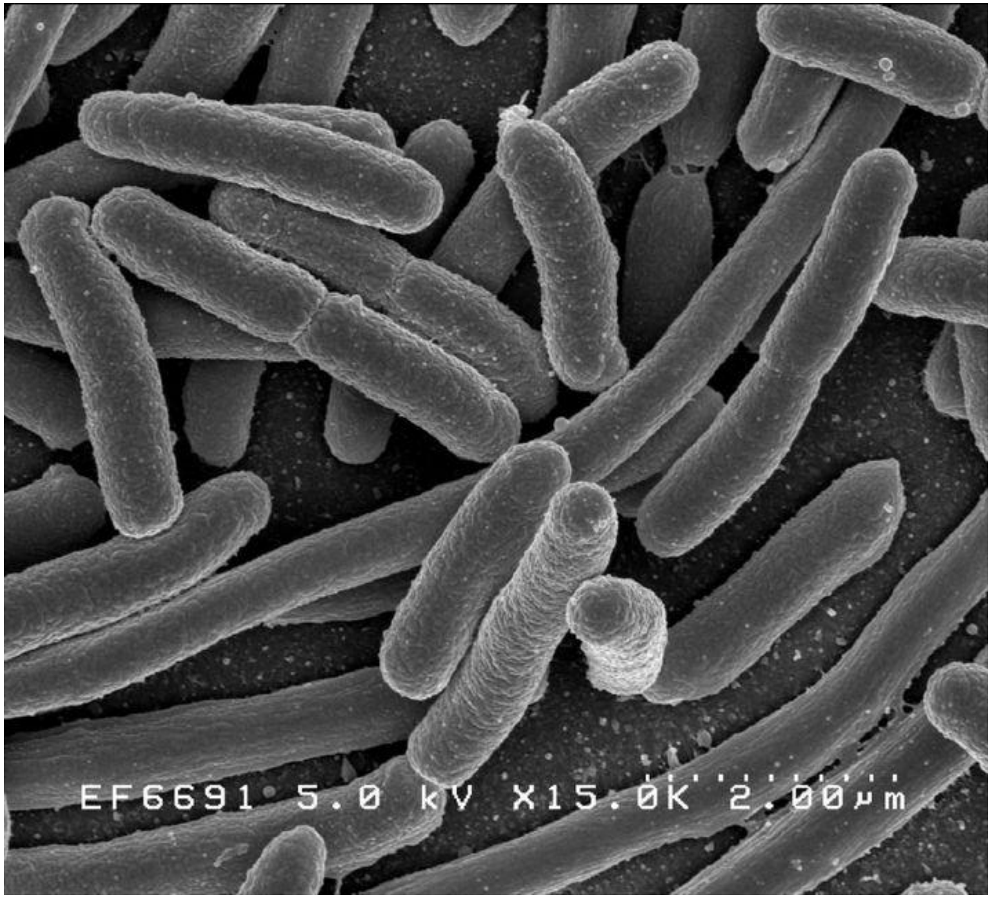


Coccus
(Kokken)

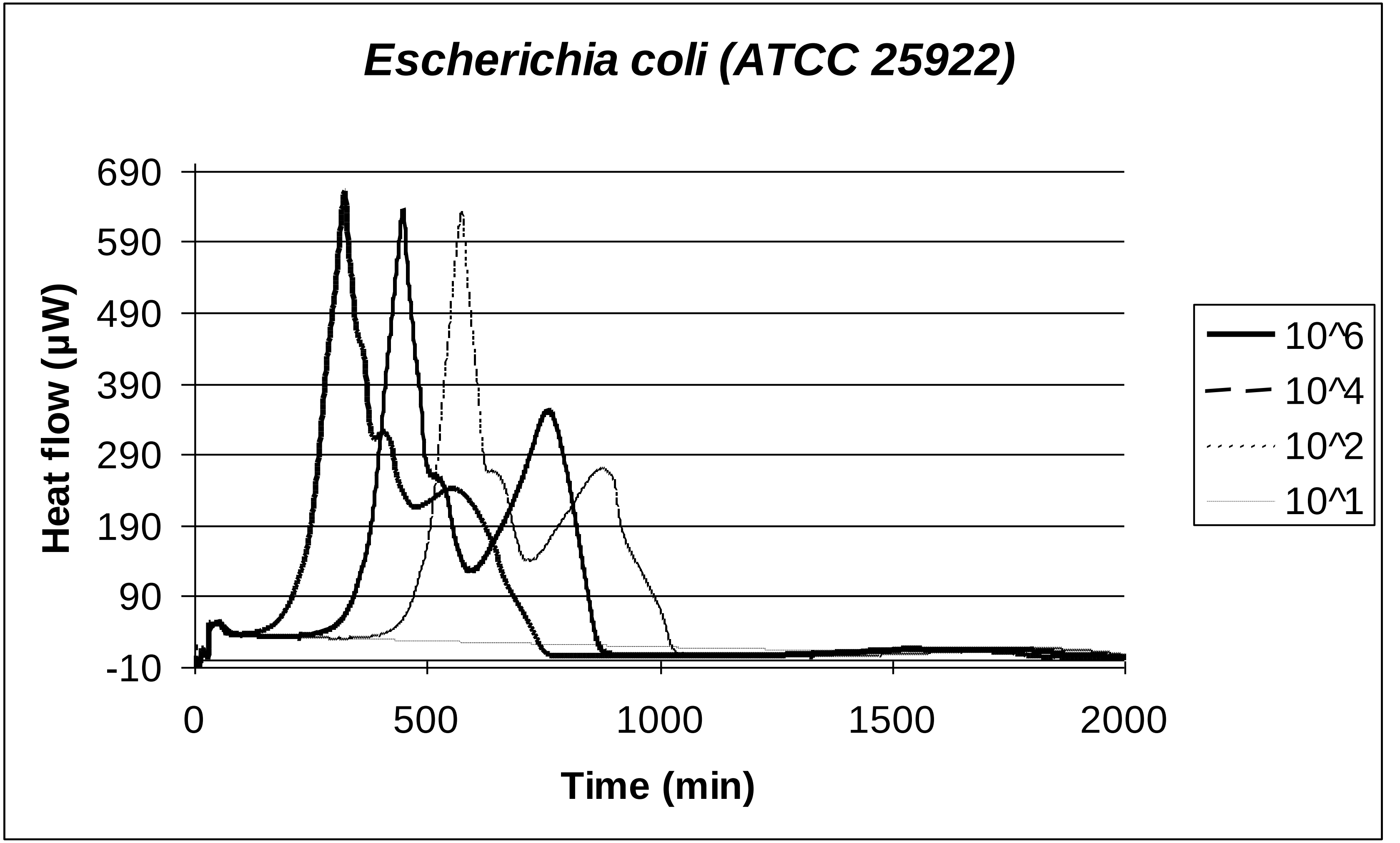
Bacili
(Stäbchen)

Spirochetes
(Spiralen)

CFU=
Colony
forming
units



Bacteria Escherichia coli, Th. Escherich (1919)



Ref. A.Trampuz et al., Biocalorimetry..., Trans-2007-0018.R2

2. Phenomenology of Bacterial Heat Production (Allometry)

Kleiber´s Law (1932)

Basic metabolic rate of aerobic living systems at T=298 K :

$$J_0 = aM^{\alpha}$$

$$a = 3{,}5 W / kg^{\alpha}$$

$$\frac{2}{3} < \alpha < 1$$

$$\alpha \cong \frac{3}{4}$$

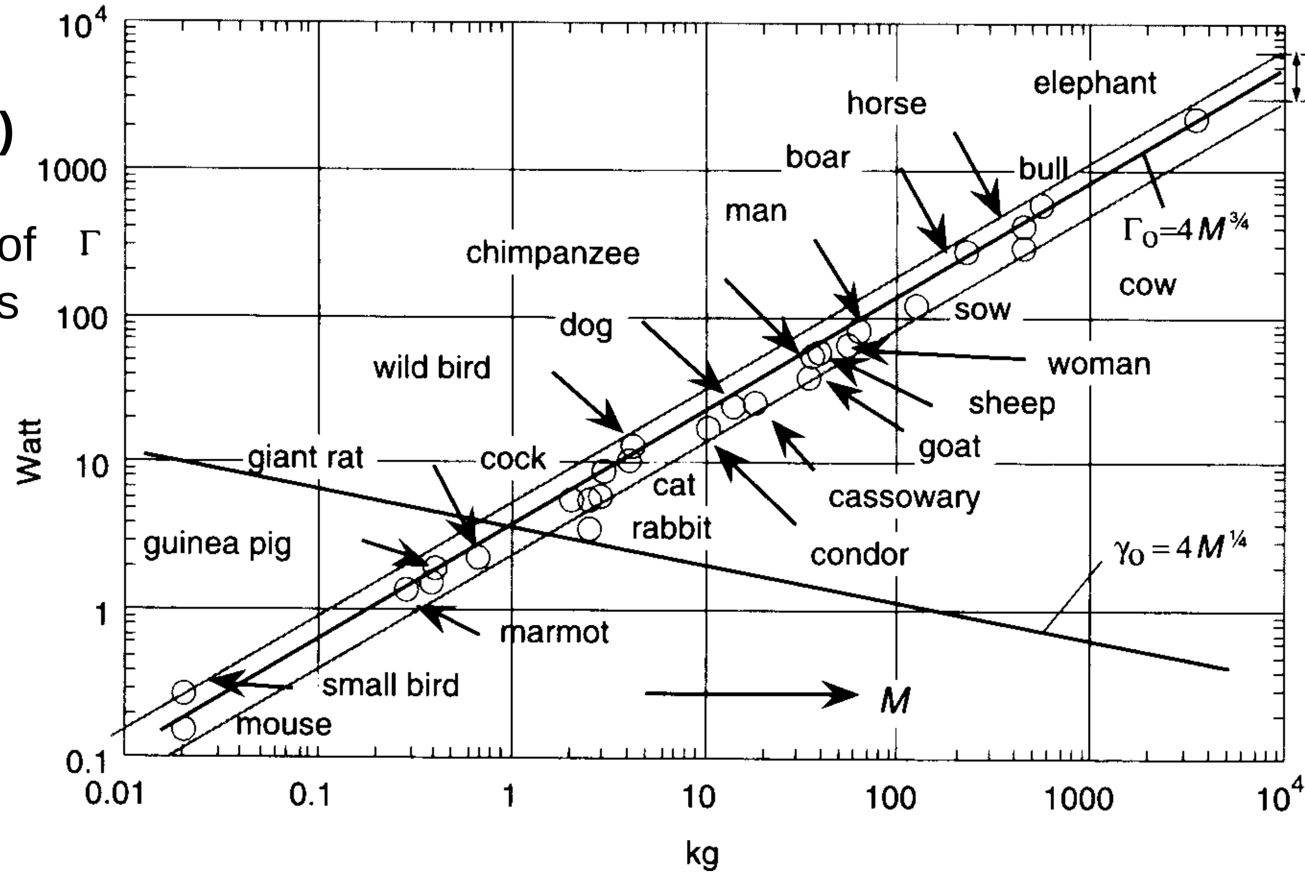
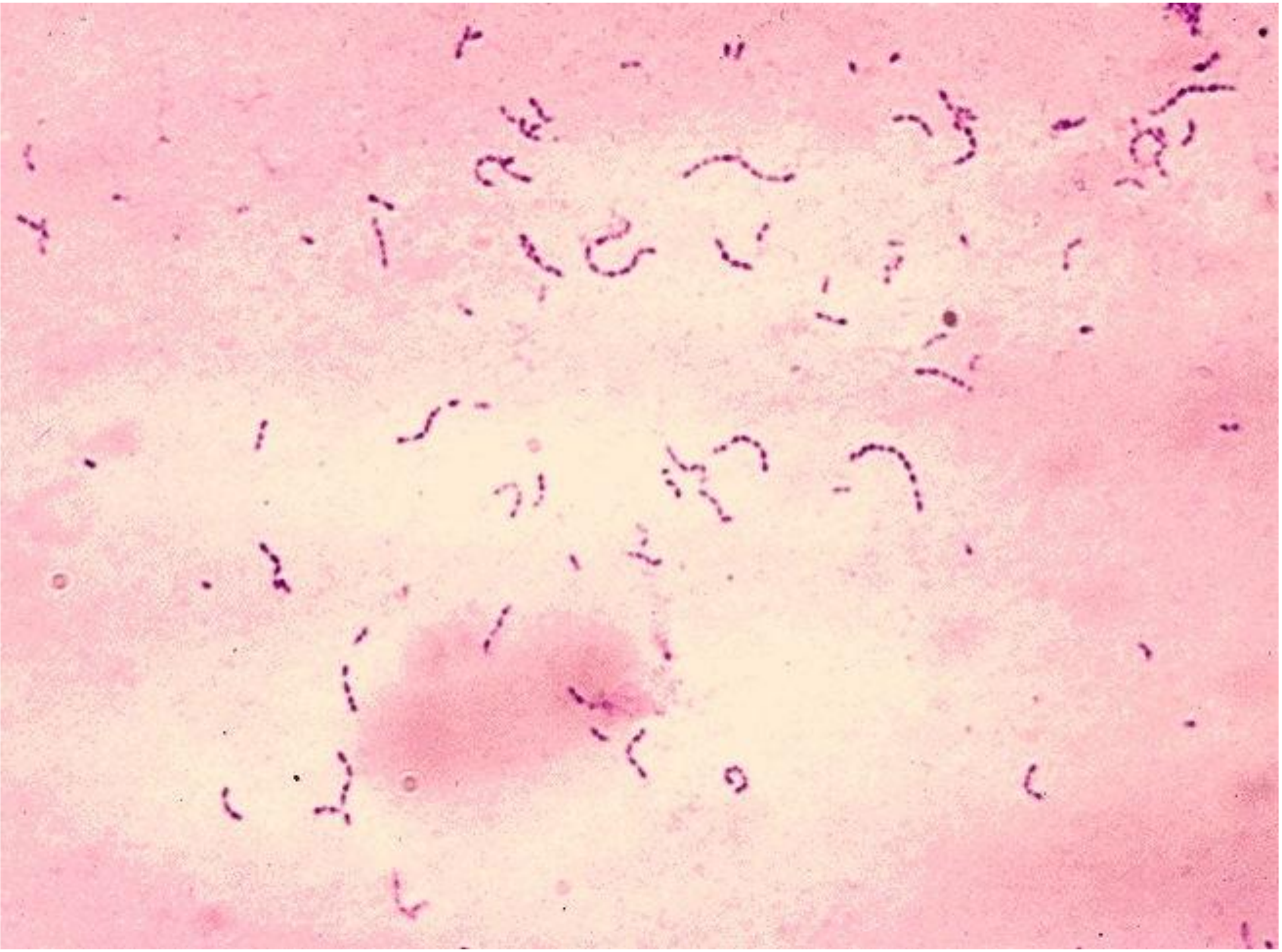
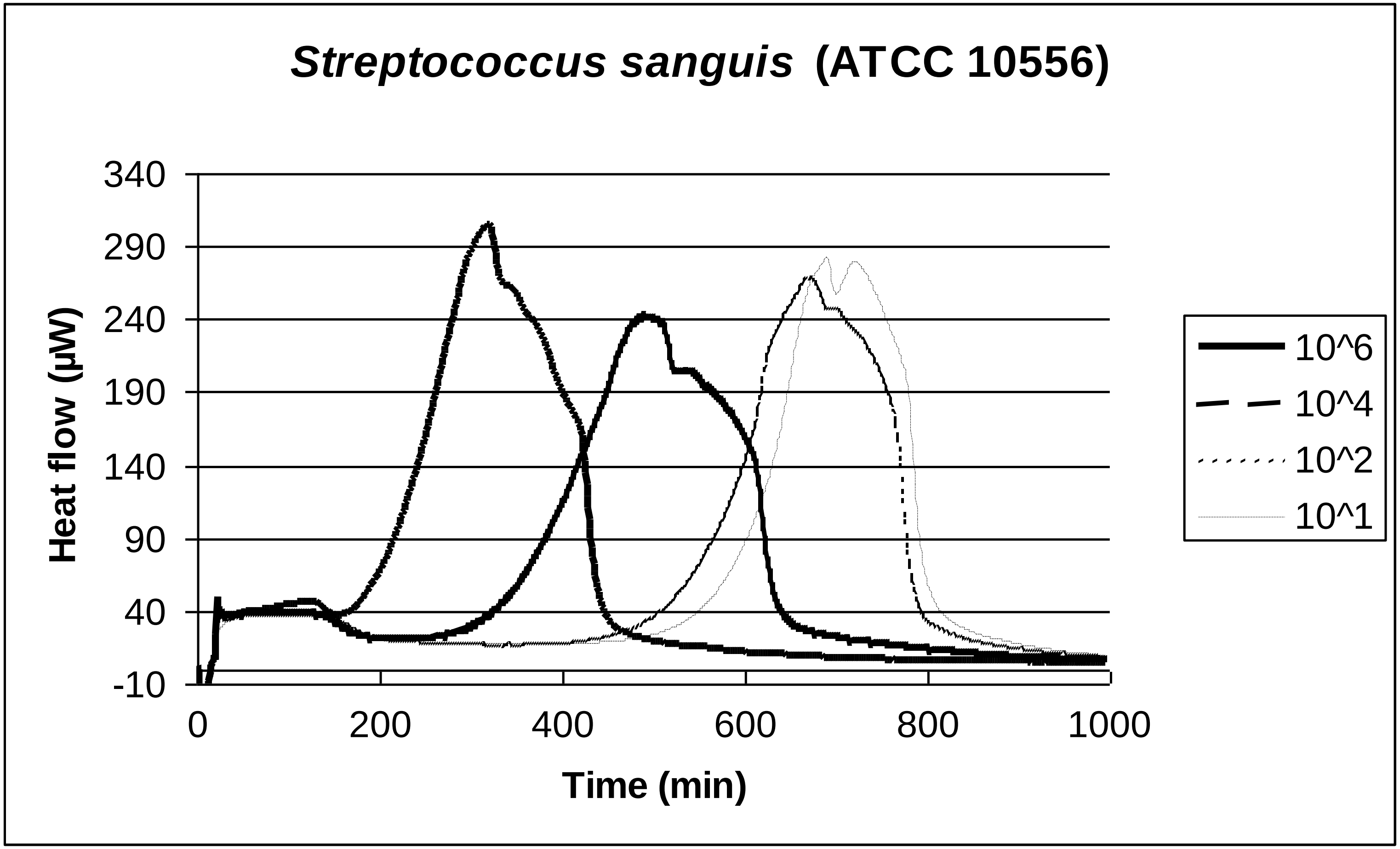


Figure A3
Metabolic rate of oxygen consumption based living systems. Mouse-Elephant-curve, Kleiber,1932. This curve also holds for bacteria (M ≅ 10* (-4) – 10*(-12) g).



Bacteria Streptococcus Mutans (Karies), Clarke (1924)



Ref. A.Trampuz et al., Biocalorimetry..., Trans-2007-0018.R2

Basic Metabolic Rate ≃ Heat Production of Aerobics

Creature	Mass/kg	Metabolic Rate J0 / W	Food Substrate	Heating Value MJ/kg	Consump-tion kg/day
Bacteria Staphylococcus aureus	$0{,}5 \bullet 10^{-15}$	120 nW	glucose	15,6	0,665. 10*(-9)
Men	80 kg	94 W	various	20	0,40
Lion	120 kg	127 W	meat	30	0,37
Elephant	3000 kg	1,418 W	grass	10	12,3

Activation factor : $J_0 \rightarrow (2\text{-}5)J_0$

Kleiber's Constant : Temperature Dependence

Bacteria growth processes, sterilisation.

$$a = a(T_b, T^*) = A.(T_B - T^*).e^{-q^*/RT^*}$$

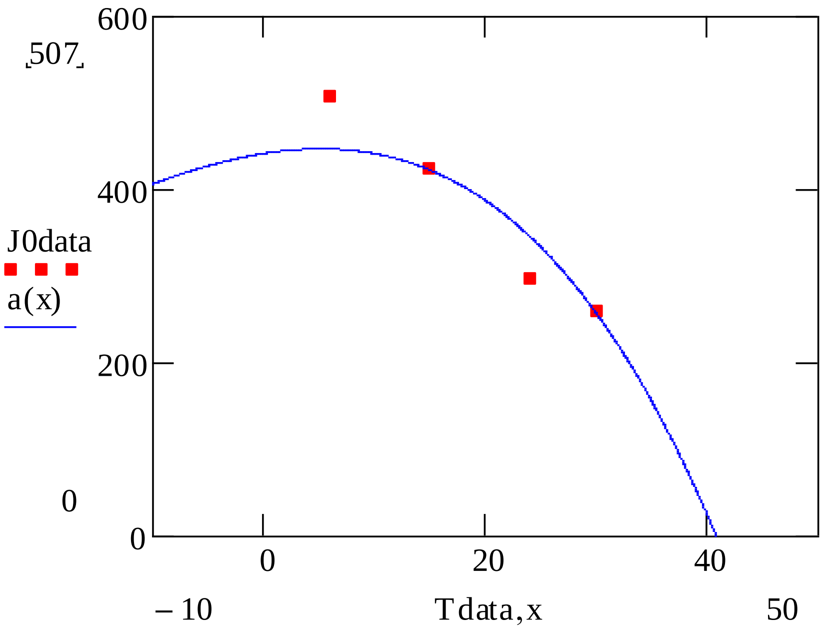
Example : Dogs,
hair cut, TB=41 C
Data : Jeroch et.al.(1999)

T_b....Maximum temperature

of living system

T^*....Environmental temperature

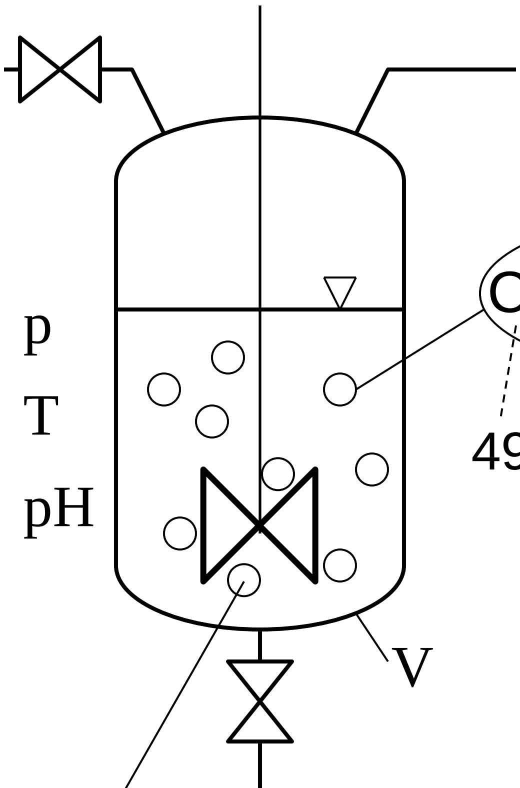
q^*....Energy (metabolism, heat transfer)



Enviromental temperature
for maximum metabolism

$$T^*_{\max} = \frac{q^*}{2R}\left(-1 + \left(1 + 4RT_b / q^*\right)^{1/2}\right)$$

Fermenter



$C_1H_{1.8}O_{0.5}N_{0.2}$

49 7.3 33 10.7 g/g

$$M \cong \frac{25g}{C-mol}$$

$$[C_x] = \frac{C-mol(x)}{e}$$ ... Biomass (x)

Example (Yeast)

Genes	5000
Metabolites	1000-5000
Concentration*)	$(0.1-10)\,m\,mol$

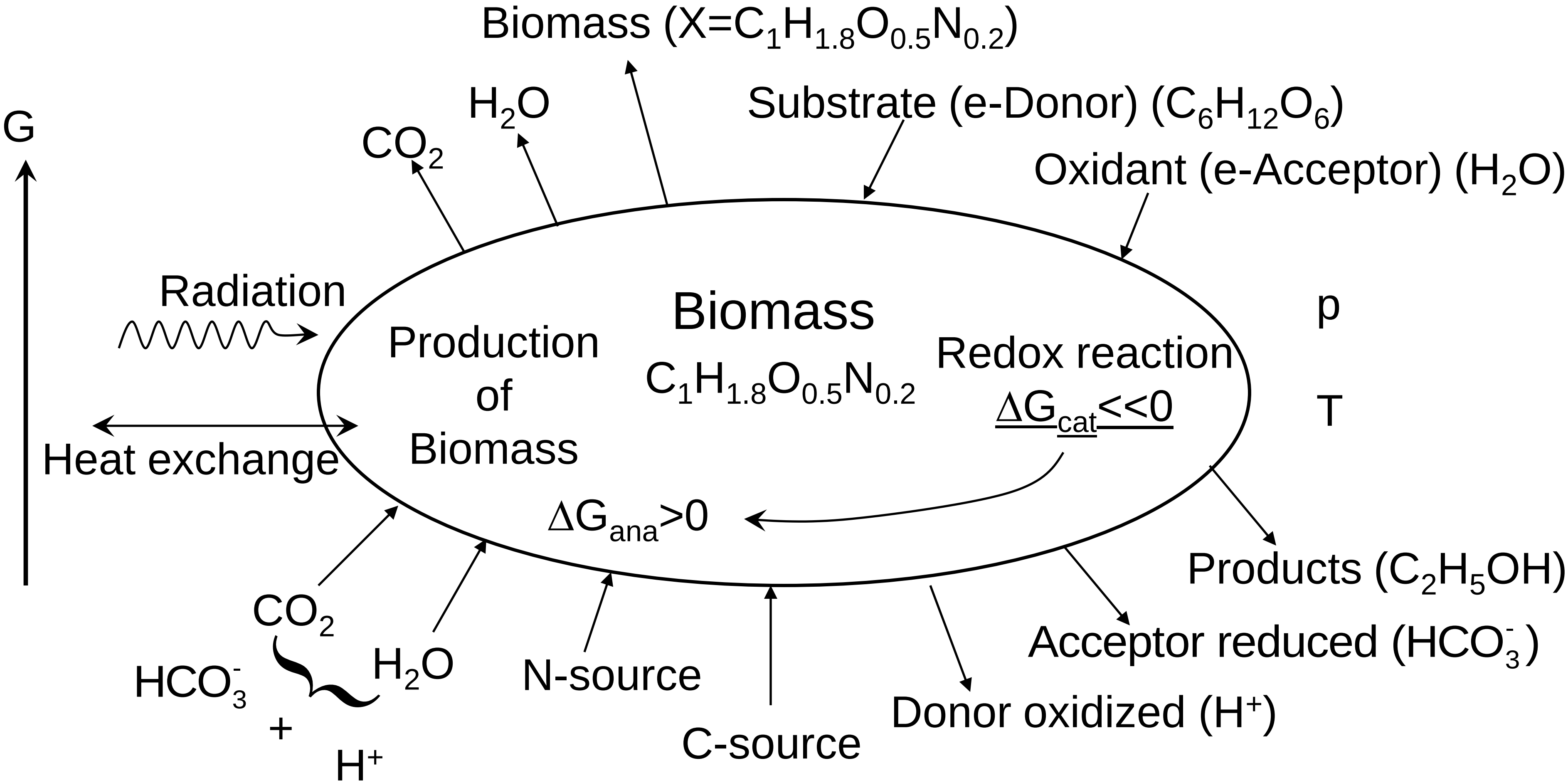
Turn over time

$$\frac{\text{Concentration}}{\text{Reaction rate}} = (1-10)s$$

*) Osmotic pressure limited. Avoiding byproducts and byreactions.

*Microbiothermodynamic system, Microbioreactor

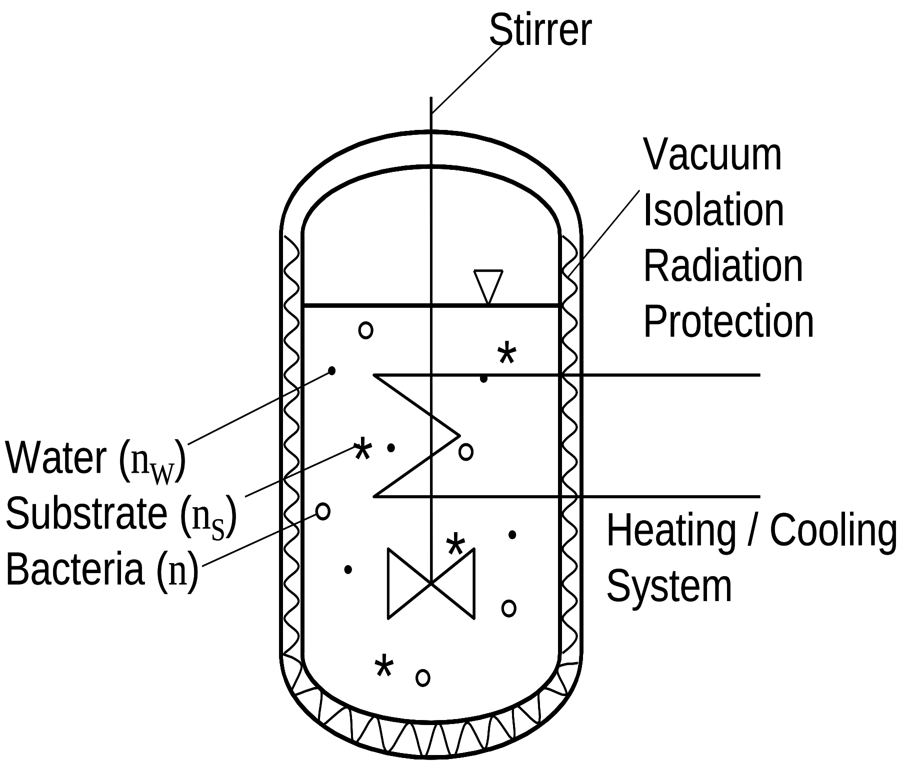
Microbial Growth System



<u>Anabolism</u> + <u>Catabolism</u> (Free Entalpy)

4. **Heat Production in Bacterial Growth Processes**

Isothermal Calorimeter



Metabolic generation of heat

$$dQ \simeq -dm_s \quad (1)$$

$$\dot{Q} = -K_s \dot{m}_s \quad (2)$$

$$Q(t) = K_s (m_{s0} - m_{s(t)}) \quad (3)$$

$$Q(\infty) = K_s m_{s0} \quad (3a)$$

Mass / molar balance growth process

$$dn \simeq -dn_s \quad (4)$$

$$\dot{n} = -C\dot{n} \quad (4a)$$

$$C \geq 0$$

Bacterial Growth Prozess Model I (Monod)

$$n(t) = n_0 + (n_\infty - n_0)\frac{(bt)^\alpha}{1+(bt)^\alpha};$$

(5)

$$n(0) = n_0 \qquad \alpha \geq 1;$$

$$n(\infty) = n_\infty \qquad b > 0.$$

Substrate

(4)→ $$\dot{n} = -C\dot{n}_s;$$

(5)→ $$n_s = \frac{n_{s0}}{1+(bt)^\alpha}.$$ (6)

Heat Production during Bacterial Growth Processes

(2)→
$$\dot{Q} = -K_s \dot{n}_s = \frac{K_s}{C} \dot{n}; \quad (7)$$

$$\dot{Q} = K_\alpha b(n_\infty - n_0) \frac{(bt)^{\alpha-1}}{(1+(bt)^\alpha)^2}. \quad (8)$$

Maximum value

$$bt_{\max} = (\alpha - 1)^{\frac{1}{\alpha}}; \quad (9)$$

$$\dot{Q}_{\max} = \frac{Kb(n_\infty - n_0)(\alpha - 1)^{1-\frac{1}{\alpha}}}{\alpha}. \quad (10)$$

Bacterial Population Growth Prozess Model II

$$dn \simeq n n_s dt \qquad (12)$$

$$\dot{n} = A n_s n \qquad (13)$$

$$n(t) = n_0 \exp\left\{ A \int_0^t n_s(t')dt' \right\} \qquad (15)$$

(3)→ $$n(t) = n_0 \exp\left\{ A \int_0^t (n_{so} - \frac{Q(t')}{(K_s)})dt' \right\} \qquad (16)$$

Bacterial Population

Heat generated in broth

Growth of Population and Depletion of Substrate

Substrate

$$dn_s \simeq -n_s n dt \qquad (17)$$

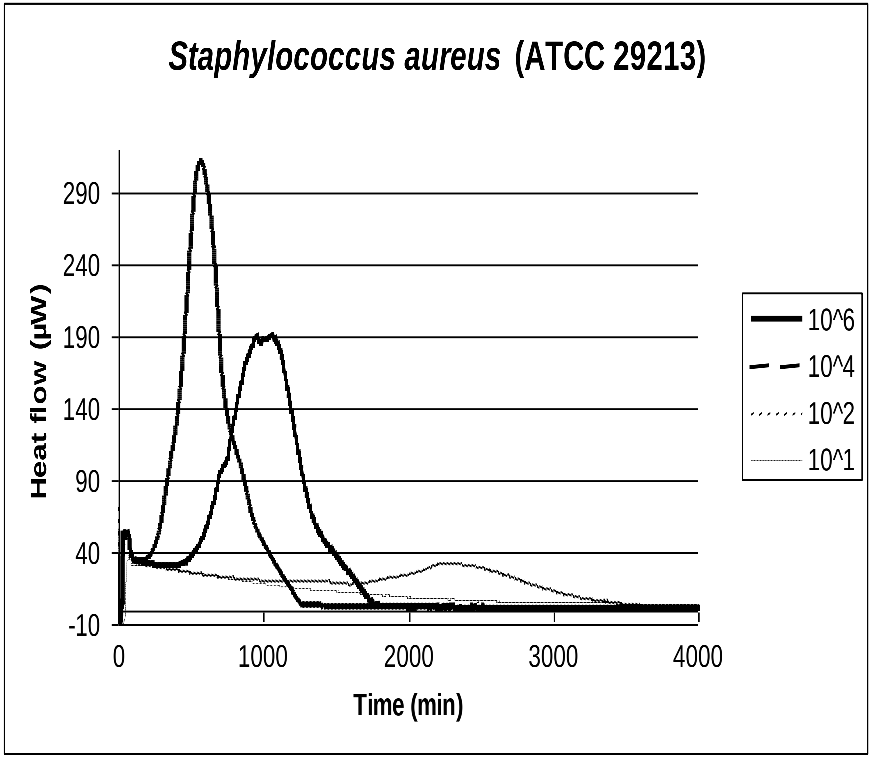
$$\dot{n} = -Bnn_s \qquad (18)$$

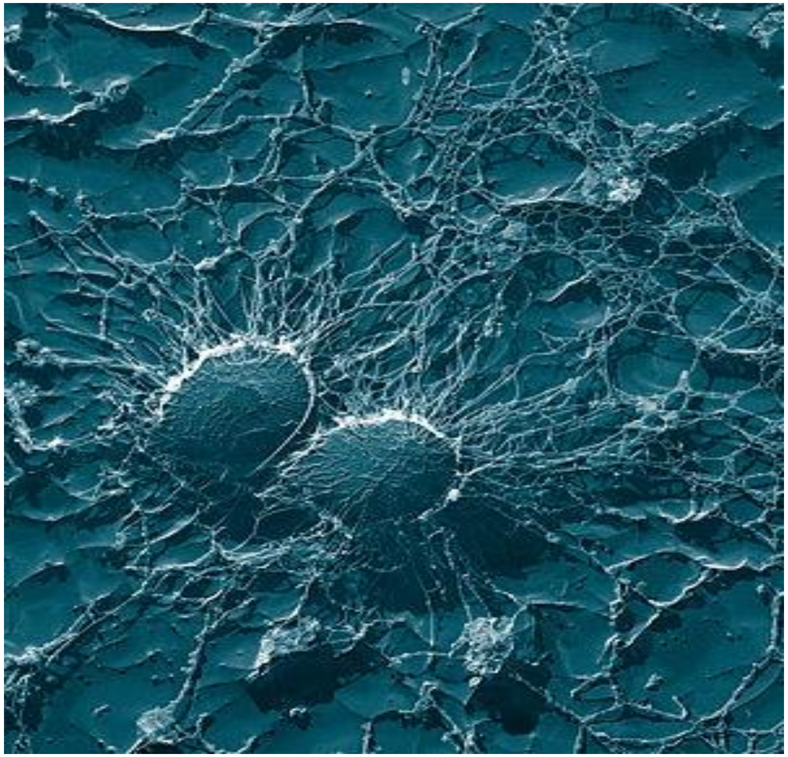
(13,18)→ $$\ddot{n} - \frac{\dot{n}^2}{n} + Bn\dot{n} = 0$$

$$\ddot{n} - \frac{\dot{n}_s{}^2}{n_s} - An_s\dot{n}_s = 0$$

$$ODEs: n = n(t),\ n_s = n_s(t)$$

Bacterial Growth Prozess Model I (Monod)





Staphylococcus aureus
(ATCC29213)

$$Size: \quad 50.000:1$$

$$Diameter: \quad (0,8-1,2)\mu m$$

$$Density: \simeq 0,8\ g/cm^3$$

Staphylococcus aureus

Population Dynamiks, Heat Production*)

$$n(t) = n_0 + (n_\infty - n_0)\frac{(bt)^\alpha}{1+(bt)^\alpha}$$

$$\dot{Q} = K\dot{n}$$

$$\dot{Q} = K\left(n_\infty - n_0\right)\alpha b \frac{(bt)^{\alpha-1}}{\left(1+(bt)^\alpha\right)^2}$$

*)Caloric measurements: Trampuz et.al., Basel

	Unit	1	2
Initial number of bacteria	CFU	10^6	10^4
Max. heat production	µW	310	190
Time at max. production	min	500	1000
Total heat generated	J	12,6	17,5
Final number bacteria	CFU	$3{,}6 \cdot 10^6$	$3{,}6 \cdot 10^6$
Time at half prod.	min.	400	800

References (Selection 1)

PLANCK M.: Vorlesung über Thermodynamik, 11. Aufl., 1964, W. de Gruyter, Berlin – New York.

VOET D & VOET J G: Biochemistry, J.Wiley&Sons,2nd Ed. 1995 New York

Lörinczy D., Editor, The Nature of Biological Systems as Revealed by Thermal Methods, Kluwer, Dordrecht, p. 355, 2004.

VON STOCKAR U., VAN DER WIELEN L.A.M., Back to Basics: Thermodynamics in Biochemical Engineering, Adv. in Biochemical Engng./Biotechnology, 80 (2003), p. 1-17.

HAINIE D.T.: Biological Thermodynamics, Cambridge University Press, Cambridge, UK, 2001.

RAFFA R.B.: Editor: Drug – Receptor Thermodyn., Introduction & Applications, J. Wiley & Sons, 2001, New York etc.

References (Selection 2)

Journal of Non-Equilibrium Thermodynamics, Review Articles,
W.de Gruyter, Berlin – New York, since 1976 :

1.WINTER R., LOPES D., GRUZIELANEK ST., VOGTT K.
Towards an Understanding of the Temperature / Pressure Configurational and Free-Energy Landscape of Biomolecules, 32(2007),p. 41 - 97.

2.HUBBUCH J., KULA M.-R.
Isolation and Purification of Biotechnological Products, 32(2007), p.99 - 127.

3.RUBI J.M., NASPREDA M., KJELSTRUP S., BEDEAUX D.
Energy Transduction in Biological Systems: A Mesoscopic Non-Equilibrium Thermodynamics Approach,
32(2007), p. 351-...

4.KELLER J.U.
An Outlook on Biothermodynamics:
Needs,Problems and New Developments
I.Stability and Hydration of Proteins, 33(2008), 321-385