THERMODYNAMICS OF BACTERIA-PHAGE INTERACTIONS
T4 and Lambda Bacteriophages, and *E. Coli* can Coexist in Natural Ecosystems Due to the Ratio of their Gibbs Energies of Biosynthesis

by

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The model of T4 phage, Lambda phage, and E. coli is often used in research on virus-host interactions. This paper reports for the first time the thermodynamic driving force of biosynthesis, catabolism and metabolism for the three organisms, on the M9 medium. Moreover, the influence of activities of nutrients and metabolic products is analyzed. All three organisms were found to have very similar Gibbs energies of metabolism. Moreover, since they share the same catabolism, their Gibbs energies of catabolism are identical. However, Gibbs energies of biosynthesis differ. The calculated thermodynamic properties have been used to explain the coexistence of both bacteria and phages in a dynamic equilibrium in natural ecosystems.

Key words: *activity, driving force, metabolism, catabolism, environment*

Introduction

Microorganisms represent biological entities, but also chemical and thermodynamic systems [1-5]. As chemical and thermodynamic systems, microorganisms interact with other microorganisms and with their environment [1, 2]. Interactions between microorganisms can be analyzed quantitatively, using chemistry and thermodynamics [4, 6-8]. Energetics of interactions between microorganisms is of great importance [9, 10]. During the last decades, a thermodynamic characterization has been made of bacteria [5, 11], viruses [3, 12-24] and phage species [24-26].

Thermodynamic methods have been applied many times in life sciences. Thermodynamic analysis of organisms is as old as thermodynamics itself. The founders of thermodynamics, Lavoisier and Laplace were the first to measure heat released by a mouse using a calorimeter [27, 28]. The first theoretical consideration was made by Boltzmann, who considered the role of entropy in energy transformations performed by organisms [29, 30]. The next was the founder of modern biothermodynamics, Schrodinger [31], who used a thermodynamic concept, entropy, to define life. Moreover, organisms were identified as open thermodynamic systems by von Bertalanffy [1, 2]. Analysis of organisms became much easier due to the work of Prigogine [32-36], who developed nonequilibrium thermodynamics as a tool that

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proved very useful in analysis of life processes. Moreover, a great contribution to the field was given by Morowitz [37-42]. More recently, von Stockar analyzed the thermodynamic driving force for life processes, identifying Gibbs energy as a convenient parameter for quantitative analysis of microbial multiplication [4, 6, 43-48]. Hansen discussed change in information content and entropy in organisms during life processes, including evolution [49-51]. Lucia discussed the importance of entropy and Gibbs energy in life sciences [52, 53], and sustainability and economy [54-56]. Calorimetric measurements and biothermodynamic analysis of microorganisms have been performed by Maskow [57-63]. Thermodynamics has been applied to the fields of soil research by Barros [64-68]. Therefore, thermodynamics has been in use in biological research for over 200 years and has given many interesting results [69].

Thermodynamics has been applied to research on viruses. Gale made thermodynamic analysis of virus-host interactions during virus attachment and entry into the host cell [70-73]. Lucia applied thermodynamics in studies on virology and epidemiology [74-77]. Maskow *et al*. [24] used calorimetry to study transitioning of viruses from lysogenic into lytic cycle. Guosheng *et al*. [25] made a calorimetric study of the bacteriophage lytic life cycle.

Calorimetry has been used to study viruses, since the accurate methods became available 50 years ago [78, 79]. Recent calorimetric studies of bacteriophages include: capsid stability [80, 81], infectivity [81-83], structure [80-84], preservation [85-89], interaction with bacteria [85, 90-93], effect against biofilms [91, 92, 94-98], capsid disassembly [99], medical applications [86-89, 100-102], transition from lysogenic to lytic cycle [24, 103], viral enzyme studies [104-106], and application in biosensors [107]. The methods used for calorimetric studies of bacteriophages include isothermal titration calorimetry, differential scanning calorimetry, and reaction calorimetry (*a.k.a*. isothermal microcalorimetry) [78, 79].

Bacteria and phages interact at two main sites: at the cell membrane and in the cytoplasm. At the cell membrane, the phages attach to the host cells and enter the cytoplasm. In the cytoplasm, the phages multiply. In both processes, energetics plays an important role [13-15, 70-73, 108].

Thermodynamic properties of viruses can be determined based on their elemental composition, using predictive models developed by Patel and Ericson [109] and Battley [110-113]. These methods are very important, since most thermodynamic laboratories lack the required biosafety level for virus analysis [16]. To enable calculation of thermodynamic properties from virus elemental composition, the atom counting method was suggested [16]. The atom counting method was found to give results in good agreement with available experimental data [3, 16].

The goal of this paper is to calculate and compare the thermodynamic driving forces for multiplication of T4 and Lambda bacteriophages, and their host *E. coli*. The analysis was performed on the M9 medium, since it is widely used and chemically well characterized. The thermodynamic driving force is quantified by Gibbs energy [6, 43], which was determined for biosynthesis, catabolism, and metabolism as a whole, for each analyzed organism. Moreover, the influence of activities of nutrients and products of microbial growth were taken into consideration.

Methods

Based on the literature data on elemental composition and standard thermodynamic properties of the T4 phage, Lambda phage, *E. coli* live matter and M9 growth medium, a thermodynamic analysis was made of the interactions between the organisms. This section describes the data sources and the approach used for the analysis.

Data sources

Elemental compositions of T4 and Lambda bacteriophages were taken from [26]. Elemental composition of the host *E. coli* was taken from [113, 114]. Elemental compositions of the analyzed organisms are given in tab. 1.

Table 1. Elemental composition of T4 and Lambda bacteriophages, and their host *E. coli*; the amount of element *J* in the empirical formula of live matter is denoted with n_J

Organism	n_C	n_H	n_O	n_N	n_P	n_S	Ref.
T4 phage	1	1.4445	0.4167	0.3120	0.0398	0.0032	[26]
Lambda phage	1	1.4174	0.4470	0.3271	0.0511	0.0031	[26]
E. coli	1	1.77	0.49	0.24	0	0	[114]
E. coli	1	1.74	0.464	0.26	0	0	[113]

Chemical composition of the M9 growth medium supplemented with glucose is given in tab. 2. The concentrations of the nutrients were taken from [115, 116]. The oxygen concentration was taken from [117]. Standard thermodynamic properties of the medium components were taken from [118-120]. Ionic radii were taken from [121, 122].

Standard thermodynamic properties of the analyzed organisms were taken from [5, 26], including standard enthalpy of formation, $\Delta_f H^0(Bio)$, standard molar entropy, $S_m{}^0(Bio)$, and standard Gibbs energy of formation, $\Delta_f G^0(Bio)$. They are given in tab. 3.

Table 2. Composition of the growth medium; the microorganisms were cultured in an M9 medium with glucose

Substance	C [mol/dm³]	a_{ion} [cm]	z [q_e]	γ_{DH}	a_{DH}
$C_6H_{12}O_6$ (aq)	0.00002397	N.A.	0	1	1
NH_4^+ (aq)	0.00935	3.00E-08	+1	0.722876	0.006759
HPO_4^{2-} (aq)	0.0337	4.00E-08	–2	0.304436	0.01026
SO_4^{2-} (aq)	0.001	4.00E-08	–2	0.304436	0.000304
O_2 (g)	2.56E-04	N.A.	0	1	0.000256
Bio	1	N.A.	0	1	1
$H_2PO_4^-$ (aq)	0.022	4.00E-08	–1	0.742804	0.016342
H_2O (l)	1	N.A.	0	1	1
Na^+ (aq)	0.07595	4.00E-08	+1	0.742804	0.056416
K^+ (aq)	0.022	3.00E-08	+1	0.722876	0.015903
Cl^- (aq)	0.0185	3.00E-08	–1	0.722876	0.013373
Mg^{2+} (aq)	0.001	8.00E-08	+2	0.410357	0.00041
Ca^{2+} (aq)	0.0003	6.00E-08	+2	0.361107	0.000108
CO_2 (aq)	1.29E-05	N.A.	0	1	1.29E-05

Stoichiometry and standard thermodynamic properties of biosynthesis

Elemental composition of the analyzed microorganisms was used to construct their biosynthesis half-reactions, macrochemical equations describing conversion of nutrients into new live matter. Elemental composition of live matter can be used to find stoichiometric coefficients in the biosynthesis half-reaction [6, 8]. This is done through stoichiometry, using conservation of matter and charge [6, 8]. The stoichiometric coefficients of the biosynthesis half-reactions were calculated using the formulas in tab. 4.

Table 3. Standard thermodynamic properties of live matter of the T4 and Lambda bacteriophages, and their host *E. coli*; the values were taken from [5]

Organism	$\Delta_f H^0$(Bio) [kJ/C-mol]	$S_m{}^0$(Bio) [J/C-molK]	$\Delta_f G^0$(Bio) [kJ/C-mol]
T4 phage	–94.4	32.6	–52.2
Lambda phage	–102.5	33.2	–59.5
E. coli	–114.1	36.4	–67.0
E. coli	–107.4	35.8	–60.9

Table 4. Stoichiometric coefficients for the biosynthesis half-reactions on the M9 growth medium; the general growth reaction on the M9 growth medium is $C_6H_{12}O_6 + NH_4^+ + HPO_4^{2-} + SO_4^{2-} + O_2 \rightarrow$ (Bio) + $H_2PO_4^- + H_2O$

Nutrient	Stoichiometric coefficient for biosynthesis half-reaction
$C_6H_{12}O_6$ (aq)	$-n_C/6$
NH_4^+ (aq)	$-n_N$
HPO_4^{2-} (aq)	$n_P + 2n_S - n_N$
SO_4^{2-} (aq)	$-n_S$
O_2 (g)	$-¼(2n_O + 3n_N - n_H - 5n_P - 6n_S)$
Bio	+1
$H_2PO_4^-$ (aq)	$n_N - 2n_P - 2n_S$
H_2O (l)	$[½(2n_O + 3n_N - n_H - 5n_P - 6n_S) + n_C + 4n_P + 4n_S - n_O]$

Standard thermodynamic properties of the biosynthesis half-reaction include standard enthalpy of biosynthesis, $\Delta_{bs}H^0$, standard entropy of biosynthesis, $\Delta_{bs}S^0$, and standard Gibbs energy of biosynthesis, $\Delta_{bs}G^0$. Standard thermodynamic properties of biosynthesis are at standard conditions: standard temperature of 298.15 K (25 °C) and pressure 1 bar, as well as unit activities of all reactants and products [118, 119]. The values of standard thermodynamic properties of biosynthesis were calculated applying the Hess's law to the biosynthesis half-reactions:

$$\Delta_r H^0 = \sum_i \nu_i \Delta_f H^0(i) \tag{1}$$

$$\Delta_r S^0 = \sum_i \nu_i S_m^0(i) \tag{2}$$

$$\Delta_r G^0 = \sum_i \nu_i \Delta_f G^0(i) \tag{3}$$

where $\Delta_r H^0$ denotes standard reaction enthalpy, $\Delta_r S^0$ standard reaction entropy, $\Delta_r G^0$ standard reaction Gibbs energy, ν_i is the stoichiometric coefficient of substance i, $\Delta_f H^0(i)$ standard enthalpy of formation of substance i, $S_m{}^0(i)$ standard molar entropy of substance I, and $\Delta_f G^0(i)$ standard Gibbs energy of formation of substance i [118, 119].

Stoichiometry and thermodynamic properties of catabolism

The energy source for all the analyzed organisms is glucose and their metabolism is aerobic. Thus, the catabolic half-reaction represents aerobic oxidation of glucose: $C_6H_{12}O_6$ + 6 O_2 = 6 CO_2 + 6 H_2O. The stoichiometric coefficients of the catabolic half-reaction were substituted into the Hess's law, eqs. (1)-(3), to find standard thermodynamic properties of catabolism, including standard enthalpy of catabolism, $\Delta_{cat}H^0$, standard entropy of catabolism, $\Delta_{cat}S^0$, and standard Gibbs energy of catabolism, $\Delta_{cat}G^0$. Since viruses hijack the meta-

bolic machinery of their host cells, they share the same catabolism [123]. Thus, the catabolic half-reaction is identical for all the analyzed microorganisms [123].

Driving force of growth and biomass yield

The catabolic and biosynthesis half-reactions are combined into the growth reaction for an organism, describing the organism's entire metabolism [4, 6, 124]. The Gibbs energy change of the growth reaction is Gibbs energy of growth (metabolism), also known as the driving force of growth [4, 6, 43]. Gibbs energy of growth of heterotrophic organisms depends only on the carbon and energy source, and not on the nature of catabolic reaction and the electron acceptor used [124]. Moreover, Gibbs energy of growth can be determined from the properties of the carbon and energy source.

Heijnen and van Dijken [125] found that the standard Gibbs energy of growth (metabolism), $\Delta_{met}G^0$, is given:

$$-\Delta_{met}G^0 = 200 + 18\left(6 - n_{C,s}\right)^{1.8} + \exp\left\{\left[\left(3.8 - \Gamma_s\right)^2\right]^{0.16}\left(3.6 + 0.4\,n_{C,s}\right)\right\} \tag{4a}$$

where $n_{C,s}$ is the number of carbon atoms in the substrate molecule (for glucose $n_{C,s} = 6$) while Γ_s denotes the degree of reduction of the substrate molecules [125]. The generalized degree of reduction of organic substance i, Γ_i, is given:

$$\Gamma_i = \frac{4\,n_{C,i} + n_{H,i} - 2\,n_{O,i}}{n_{C,i}} \tag{4b}$$

where $n_{C,i}$, $n_{H,i}$ and $n_{O,i}$ are the numbers of C, H, and O atoms in the formula of the substance, respectively [8]. Equation (4a) was developed empirically by considering growth yields of many microorganisms on various substrates [125]. The explanation is that there is an optimum substrate degree of reduction, which is similar to that of the live matter [124, 125]. At this degree of reduction, Gibbs energy required to drive the metabolism is minimal, since less changes need to be made to the substrate to incorporate it into the organism's live matter [124, 125]. Going away from the optimal degree of reduction means that more Gibbs energy has to be dissipated [124, 125].

A similar equation was proposed by Liu *et al*. [126]:

$$\Delta_{met}G^0 = \frac{666.2}{\Gamma_s} + 243.1 \quad \text{for} \quad \Gamma_s \le 4.67 \tag{5a}$$

$$\Delta_{met}G^0 = 157\Gamma_s - 339 \quad \text{for} \quad \Gamma_s > 4.67 \tag{5b}$$

This equation is based on the considerations of Roels about biomass yields [127]. The third solution is to approximate the standard Gibbs energy of growth with that of most microorganisms, $\Delta_{met}G^0 = -500$ kJ/C-mol [6]. All three approaches were used in this research.

Gibbs energy of growth was used to find biomass yield, Y, using the equation [6]:

$$Y = \frac{\Delta_{cat}G^0}{\Delta_{met}G^0 - \Delta_{bs}G^0} \tag{6}$$

Standard thermodynamic properties of growth

The biomass yield can be used to combine the catabolic and biosynthesis half-reactions into the full growth reaction [6]. The catabolic and biosynthesis half-reactions are two parts of the total metabolism [6]. They are combined using the biomass yield [6]. The catabolic

half-reaction is divided with the biomass yield and added to the biosynthesis half-reaction. Thus, the stoichiometric coefficient of substance *i* in the total growth reaction, $\nu_{i,met}$, is:

$$\nu_{i,met} = \frac{1}{Y}\nu_{i,cat} + \nu_{i,bs} \tag{7}$$

where $\nu_{i,cat}$ and $\nu_{i,bs}$ are stoichiometric coefficients of substance *i* in the catabolic and biosynthesis half-reactions, respectively [6].

Similarly, standard thermodynamic parameters of the growth reaction are found by combining those of the catabolic and biosynthesis half-reactions [6]. Standard enthalpy, $\Delta_{met}H^0$, and entropy, $\Delta_{met}S^0$, of growth (metabolism) are given [6]:

$$\Delta_{met}H^0 = \frac{1}{Y}\Delta_{cat}H^0 + \Delta_{bs}H^0 \tag{8}$$

$$\Delta_{met}S^0 = \frac{1}{Y}\Delta_{cat}S^0 + \Delta_{bs}S^0 \tag{9}$$

Standard Gibbs energy of growth is calculated using predictive models, as described in the section *Driving force of growth and biomass yield*.

Activities

All thermodynamic parameters discussed thus far are under standard conditions: standard temperature of 298.15 K (25 °C) and pressure 1 bar, as well as unit activities of all reactants and products [118, 119]. However, it would be good to include the influence concentrations and activities of the reactions and products. The influence of activity on reaction Gibbs energy, $\Delta_r G$, is given:

$$\Delta_r G = \Delta_r G^0 + \mathrm{R}T\ln Q \tag{10}$$

where $\Delta_r G^0$ is standard reaction Gibbs energy, R the universal gas constant, *T* the temperature, and *Q* the reaction quotient [118, 119]. The reaction quotient is defined through the activities and stoichiometric coefficients of the reaction participants [118, 119]:

$$Q = \prod_i a_i^{\nu_i} \tag{11}$$

where ν_i and a_i are the stoichiometric coefficient and activity of substance *i*, respectively [118, 119]. Activity of substance *i* is given:

$$a_i = \gamma_i C_i \tag{12}$$

where γ_i and C_i are the activity coefficient and molarity of substance *i*, respectively [118, 119].

The problem of activities has been approached in four ways. The first and easiest method is to set all activities to 1. Substituting this into eq. (11) gives $Q = 1$. This means that the R*T* ln*Q* term from eq. (10) becomes zero. Thus, the reaction Gibbs energy, $\Delta_r G$, reduces to the standard reaction Gibbs energy, $\Delta_r G^0$. In other words, the influence of activities on reaction Gibbs energy is assumed to be very small and hence negligible. This modifies eqs. (10)-(12) into:

$$\Delta_r G \approx \Delta_r G^0 \tag{13a}$$

$$Q = 1 \tag{13b}$$

$$a_i = 1 \text{ for all } i \tag{13c}$$

In the present discussion, this means approximating Gibbs energies of biological processes $\Delta_{cat}G$, $\Delta_{bs}G$, and $\Delta_{met}G$ with their standard values $\Delta_{cat}G^0$, $\Delta_{bs}G^0$, and $\Delta_{met}G^0$.

The second approach is to approximate activities of all the components with their concentrations, tab. 2. This means that γ_i is set to 1 for all the considered substances in eq. (12), leaving $a_i = C_i$ [118, 119, 122]. Substituting this into eq. (11) gives the concentration reaction quotient, Q_C [118, 119, 122]. The concentration reaction quotient is then substituted into eq. (10) to give reaction Gibbs energy with corrections for concentrations, $\Delta_r G_C$. This means that eqs. (10)-(12) become:

$$\Delta_r G_C = \Delta_r G^0 + \mathrm{R}T \ln Q_C \tag{14a}$$

$$Q_C = \prod_i C_i^{\nu_i} \tag{14b}$$

$$a_i \approx C_i \quad \text{for} \quad \text{all } i \tag{14c}$$

Approximating activities with concentrations means assuming intermolecular forces between molecules in the solution are not very strong and can be neglected [118, 119, 122]. This assumption was used to calculate Gibbs energy of biosynthesis with corrections for concentrations, $\Delta_{bs}G_C$, which will later be compared to the values calculated using the other three approaches.

The third approach is to include only the activity coefficients of ions, which interact the strongest, using the extended Debye-Huckel equation. The assumption that there are no intermolecular forces in the solution is the least accurate for ions [118, 119, 122]. Ions are charged and hence interact through coulomb forces, which are strong and have a relatively long range [118, 119, 122]. These interactions are taken into account by the activity coefficients, which can be calculated using the extended Debye-Huckel equation [60, 128]. According to the extended Debye-Huckel equation, the activity coefficients depend on the ionic force of the solution, I, given:

$$I = \frac{1}{2}\sum_i C_i z_i^2 \tag{15}$$

where C_i and z_i are the concentration (molarity) and charge of species i, respectively. The sum is over all the species in the solution [60, 118, 119, 122, 128]. The ionic force is then used to calculate the activity coefficient of each species, using the extended Debye-Huckel equation:

$$\log_{10}(\gamma_i) = -\frac{A z_i^2 \sqrt{I}}{1 + B a_{ion,i} \sqrt{I}} \tag{16}$$

where A and B are constants, while $a_{ion,i}$ represents the effective diameter of the hydrated ion i [60, 118, 119, 122, 128]. For aqueous solutions at 25 °C, A = 0.5085 and B = 3.3 Å^{-1} [122]. Once the values of γ_i are substituted into eq. (12), the Debye-Huckel activates are obtained, which are then used in eq. (11) to find the Debye-Huckel reaction quotient, Q_{DH}. The Debye-Huckel reaction quotient is substituted into eq. (10) to find the reaction Gibbs energy with Debye-Huckel activities, $\Delta_r G_{DH}$. This approach was used to find $\Delta_{bs}G_{DH}$ values.

The fourth and most accurate method is to include activities of uncharged species, such as glucose, in addition to the Debye-Huckel activities for ions. The activity correction for glucose, which is not charged, was calculated as described in [129]. Standard Gibbs energy of formation of glucose *in an aqueous solution* is equal to its standard Gibbs energy of

formation *in solid state* plus two corrections: solution and dilution. The correction for solution was taken from [129]. The dilution correction was made using the Margules model, as described in [129]. Once the dilution correction is taken into account, thermodynamic parameters of glucose in an 23.97 μM solution are $\Delta_f H^0$(glc, aq) = –1265.84 kJ/mol, $S_m{}^0$ (glc, aq) = 250.35 J/mol K, and $\Delta_f G^0$(glc, aq) = –921.15 kJ/mol. These values are then used to find $\Delta_r G^0$*, reaction Gibbs energy with the glucose correction. The $\Delta_r G^0$* is then added to the reaction quotient calculated using the Debye-Huckel activities, but with the activity of glucose set to 1 (since it was already included into $\Delta_r G^0$* no further corrections are needed).

The activity of live matter was set to 1 in all four approaches, since it represents a distinct phase from the surrounding solution. Live matter is present in the solution in the form of cells (*E. coli*) or virus particles (T4 and Lambda phages). Cells are separated from the growth medium by the cell membrane and hence represent a distinct phase. Similarly, T4 and Lambda phages possess protein capsids, which separate the virus particles from the growth medium and make them a distinct phase. Thus, since both cells and virus particles represent separate phases from the growth medium, their activities are a_{bio} = 1 [118, 119]. Similarly, water is the solvent and its activity is a_{H2O} = 1 [118, 119].

Results

Equations for calculating stoichiometric coefficients for biosynthesis reactions on the M9 growth medium were formulated. They are given in tab. 4. Elemental composition of live matter was used to formulate equations for stoichiometric coefficients for microorganism biosynthesis on the M9 medium. The equations from tab. 4 were combined with the elemental compositions of live matter from tab. 1 to find the values of the stoichiometric coefficients of the analyzed organisms, given in tab. 5. The stoichiometric coefficients from tab. 5 are for the general growth reaction on the M9 growth medium:

$$C_6H_{12}O_6 + NH_4^+ + HPO_4^{2-} + SO_4^{2-} + O_2 \rightarrow (\text{Bio}) + H_2PO_4^- + H_2O \qquad (17)$$

where (Bio) denotes live mater with elemental composition from tab. 1.

Table 5. Values of the stoichiometric coefficients for the biosynthesis half-reactions of the analyzed organisms; the values were calculated using the formulas from tab. 3 and elemental composition of live matter from tab. 1, the values of stoichiometric coefficients of products are positive, while those of reactants are negative

Organism	$C_6H_{12}O_6$ (aq)	NH_4^+ (aq)	HPO_4^{2-} (aq)	SO_4^{2-} (aq)	O_2 (g)	Bio	$H_2PO_4^-$ (aq)	H_2O (l)
T4 phage	-0.1667	-0.3120	-0.26584	-0.0032	-0.0267	1	0.2260	0.8085
Lambda phage	-0.1667	-0.3271	-0.26989	-0.0031	-0.0460	1	0.2188	0.8618
E. coli	-0.1667	-0.24	-0.24	0	0.0175	1	0.24	0.475
E. coli	-0.1667	-0.26	-0.26	0	0.0080	1	0.26	0.52

Stoichiometric coefficients from tab. 5 were combined with standard thermodynamic properties of live matter from tab. 3, to find standard thermodynamic properties of biosynthesis, using the Hess's law, eqs. (1)-(3). Standard thermodynamic properties of biosynthesis include standard enthalpy of biosynthesis, $\Delta_{bs}H^0$, standard entropy of biosynthesis, $\Delta_{bs}S^0$, and standard Gibbs energy of biosynthesis, $\Delta_{bs}G^0$. Standard thermodynamic properties of biosynthesis of the T4 phage, Lambda phage, and *E. coli* live matter are given in tab. 6. Standard enthalpy of biosynthesis for *E. coli* is between –6.9 and –10.4 kJ/C-mol, while those of the T4 and Lambda phages are –18.6 kJ/C-mol and –25.4 kJ/C-mol, respectively. Standard entropy of biosynthesis of *E. coli* is between 35.3 and 35.8 J/C-mol K, while those of the T4 and Lambda phages are 35.8 and 33.6 kJ/C-mol, respectively. Standard Gibbs energy of biosyn-

thesis of *E. coli* is between –17.6 and –21.4 kJ/C-mol, while those of the T4 and Lambda phages are –29.9 kJ/C-mol and –36.1 kJ/C-mol, respectively.

Table 6. Standard thermodynamic properties of biosynthesis of the analyzed organisms

Organism	$\Delta_{bs}H^0$ [kJ/C-mol]	$\Delta_{bs}S^0$ [J/C-molK]	$\Delta_{bs}G^0$ [kJ/C-mol]
T4 phage	–18.6	35.8	–29.9
Lambda phage	–25.4	33.6	–36.1
E. coli	–6.9	35.3	–17.6
E. coli	–10.4	35.8	–21.4

Table 7. Comparison of values of Gibbs energies of biosynthesis obtained with various activity corrections

Organism	$\Delta_{bs}G^0$ [kJ/C-mol]	$\ln(Q_c)$	$\Delta_{bs}G_c$ [kJ/C-mol]	$\ln(Q_{DH})$	$\Delta_{bs}G_{DH}$ [kJ/C-mol]	$\ln(Q_a)$	$\Delta_{bs}G$ [kJ/C-mol]
T4 phage	–29.9	3.512	–21.2	3.866	–20.3	2.093	–24.07
Lambda phage	–36.1	3.783	–26.7	4.149	–25.8	2.376	–29.54
E. coli	–17.6	2.647	–11.0	2.939	–10.3	1.166	–14.04
E. coli	–21.4	2.811	–14.5	3.127	–13.7	1.354	–17.41

Standard Gibbs energies of biosynthesis were combined with the medium composition, to find corrections for activities of the reactants and products. The results are given in tab. 7. The composition of the growth medium is given in tab. 2. The activities were treated in four ways, as described in the section Activities. The first and simplest method is assuming that activities of all the substances are equal to 1. This means that they give a negligible contribution to the Gibbs energy of biosynthesis in eq. (10), giving eq. (13) and hence $\Delta_{bs}G \approx \Delta_{bs}G^0$. The second approach is to approximate activities with concentrations of substances, using eq. (14). This results in the concentration reaction quotient, Q_c, and Gibbs energy of biosynthesis with concentrations taken into account, $\Delta_{bs}G_c$, in tab. 7. The third approach is to include activity coefficients of ions only, using the extended Debye-Huckel eq. (16). The Debye-Huckel activity coefficients are substituted into eqs. (10)-(12). The result is the Debye-Huckel reaction quotient, Q_{DH}, and Debye-Huckel Gibbs energy of biosynthesis, $\Delta_{bs}G_{DH}$, values in tab. 7. The fourth approach is to include activity of glucose, in addition to that of ions, using the approach described in [129]. This results in the Q_a and $\Delta_{bs}G$ values in tab. 7.

Table 8. Stoichiometry and thermodynamic parameters of the catabolic half-reaction; for all the analyzed organisms, the catabolic half-reactions are identical, the stoichiometric coefficients of the products are positive, while those of the reactants are negative products and reactants

Stoichiometry				
$C_6H_{12}O_6$ (aq)	O_2 (g)	CO_2 (aq)	H_2O (l)	
–1	– 6	6	6	
Thermodynamic parameters				
$\Delta_{cat}H^0$ [kJ/mol]	$\Delta_{cat}S^0$ [J/mol K]	$\Delta_{cat}G^0$ [kJ/mol]	$\ln(Q_a)$	$\Delta_{cat}G$ [kJ/mol]
– 2802.74	258.97	– 2871.74	– 17.92	– 2861.93

All the analyzed organisms are growing aerobically on the M9 medium with glucose. Thus, their carbon and energy source is glucose, while the electron acceptor is oxygen. Therefore, the catabolic half-reaction is oxidation of glucose by O_2. The catabolic half-reaction and its thermodynamic properties are given in tab. 8. For every mole of glucose consumed, $\Delta_{cat}H^0$ = –2802.74 kJ/mol of heat is released. Standard entropy of catabolism is 258.97 J/mol K. Thus, the catabolic half-reaction has a highly negative standard Gibbs energy

change of –2871.74 kJ/mol. This value can be more accurate by including the correction for activities, as described in the section *Activities*. This gives the Gibbs energy of the catabolic half-reaction of –2861.93 kJ/mol.

Catabolic and biosynthesis half-reactions are combined to find the total growth reaction [6]. Gibbs energy of growth (metabolism), $\Delta_{met}G^0$, was calculated using eq. (4) by Heijnen and van Dijken [125], eq. (5) by Liu *et al*. [126], and the approximate value for most microorganisms: –500 kJ/C-mol [6]. The results are given in tab. 9. These were combined with standard Gibbs energies of catabolic and biosynthetic half-reactions to find the biomass yield, *Y*, using eq. (6).

Table 9. Substrate degree of reduction, standard Gibbs energy of metabolism and biomass yields of the analyzed organisms; $\Delta_{met}G^0$ and Y were calculated using three models: Heijnen equation [125], Liu equation [126], and approximate value for most microorganisms [6]

Organism	Substrate	Γ_S	$\Delta_{met}G^0$ (kJ/C-mol)			$\Delta_{cat}G^0$ [kJ/C-mol]	$\Delta_{bs}G^0$ [kJ/C-mol]	Y		
			Heijnen	Liu	Approx.			Heijnen	Liu	Approx.
T4 phage	$C_6H_{12}O_6$	4.0	–236.1	–409.7	–500	–2871.7	–29.9	13.9	7.6	6.1
Lambda phage	$C_6H_{12}O_6$	4.0	–236.1	–409.7	–500	–2871.7	–36.1	14.4	7.7	6.2
E. coli	$C_6H_{12}O_6$	4.0	–236.1	–409.7	–500	–2871.7	–17.6	13.1	7.3	6.0
E. coli	$C_6H_{12}O_6$	4.0	–236.1	–409.7	–500	–2871.7	–21.4	13.4	7.4	6.0

The biomass yield was used to combine the catabolic and biosynthetic half-reactions to find the total growth reaction, using eq. (7). The stoichiometric coefficients for the total growth reactions of the analyzed organisms are given in tab. 10. Moreover, the biomass yields were used to find standard enthalpy, $\Delta_{met}H^0$, and entropy, $\Delta_{met}S^0$, of growth (metabolism), using eqs. (8) and (9). Their values are shown in tab. 11. Finally, the correction for activities was made using eqs. (10)-(12), giving Gibbs energy of growth (metabolism), $\Delta_{met}G$. The values of $\Delta_{met}G$ for the analyzed organisms, along with the Gibbs energies of catabolism and biosynthesis, are shown in tab. 12.

Table 10. Stoichiometric coefficients of entire growth reactions, including both catabolism and anabolism, of the analyzed organisms

Organism	$C_6H_{12}O_6$ (aq)	NH_4^+ (aq)	HPO_4^{2-} (aq)	SO_4^{2-} (aq)	O_2 (g)	Bio	$H_2PO_4^-$ (aq)	H_2O (l)	CO_2 (aq)
T4 phage	–0.2989	–0.3120	–0.2658	–0.0032	–0.8201	1	0.2260	1.6019	0.7934
Lambda phage	–0.2967	–0.3271	–0.2699	–0.0031	–0.8265	1	0.2188	1.6422	0.7805
E. coli	–0.3032	–0.2400	–0.2400	0.0000	–0.8016	1	0.2400	1.2941	0.8191
E. coli	–0.3019	–0.2600	–0.2600	0.0000	–0.8031	1	0.2600	1.3311	0.8111

Table 11. Standard thermodynamic properties of entire metabolic reactions of the analyzed organisms

Organism	$\Delta_{met}H^0$ [kJ/C-mol]	$\Delta_{met}S^0$ [J/C-mol K]	$\Delta_{met}G^0$ [kJ/C-mol]
T4 phage	–389.2	70.0	–409.7
Lambda phage	–390.0	67.3	–409.7
E. coli	–389.5	70.6	–409.7
E. coli	–389.3	70.8	–409.7

Discussion

This paper analyzes the interaction of *E. coli* with T4 and Lambda bacteriophages, on the M9 growth medium with glucose. Thus, elemental compositions and standard thermodynamic properties of the analyzed microorganisms were taken from the literature [5, 26, 113,

Table 12. Gibbs energies of catabolism, biosynthesis and entire metabolism of the analyzed microorganisms, including the activities of the nutrients and metabolic products

Organism	$\Delta_{cat}G$ [kJ/C-mol]	$\Delta_{bs}G$ [kJ/C-mol]	$\Delta_{met}G$ [kJ/C-mol]
T4 phage	–2861.93	–24.07	–402.5
Lambda phage	–2861.93	–29.54	–401.8
E. coli	–2861.93	–14.04	–404.8
E. coli	–2861.93	–17.41	–404.3

114]. The composition of the growth medium was also taken from the literature [115-117], as well as the thermodynamic properties of its constituents [118-122]. These were combined to find the catabolic and biosynthetic half-reactions for *E. coli*, and T4 and Lambda phages. Moreover, standard thermodynamic properties of catabolism and biosynthesis were calculated, as well as thermodynamic properties with corrections for activities of substances in the growth medium. Finally, the driving force of growth and complete growth reactions were formulated for the analyzed microorganisms.

Biosynthesis

Table 5 gives the stoichiometric coefficients for the biosynthesis half-reactions for the analyzed microorganisms. The stoichiometric values for O_2 are different for the bacteriophages and the host cells. For the bacteriophages they are negative, meaning oxygen is consumed for biosynthesis, in excess to that used in catabolism [6, 124]. On the other hand, for *E. coli* they are positive, meaning that no additional oxygen is used for biosynthesis [6, 124]. The reason for this is the higher nitrogen content of viruses. The consumption of O_2 in biosynthesis of T4 and Lambda phages means that the substrate is partly oxidized while it is incorporated into phage live matter. The oxidation process is exothermic. Thus, the two bacteriophages have more negative standard enthalpies of biosynthesis, tab. 6. The T4 phage has a standard enthalpy of biosynthesis of –18.6 kJ/C-mol, for Lambda phage it is –25.4 kJ/C-mol, while for *E. coli* $\Delta_{bs}H^0$ is between –6.9 and –10.4 kJ/C-mol. Moreover, the more negative enthalpies make standard Gibbs energies of biosynthesis of the T4 and Lambda phages more negative that of their host *E. coli*, according to the equation $\Delta_{bs}G^0 = \Delta_{bs}H^0 - T\Delta_{bs}S^0$. Thus, for the T4 phage standard Gibbs energy of biosynthesis is –29.9 kJ/C-mol, for the Lambda phage it is –36.1 kJ/C-mol, while for *E. coli* $\Delta_{bs}G^0$ is between –17.6 and –21.4 kJ/C-mol, tab. 6. Therefore, the chemical structure of phages is reflected in their elemental composition. The elemental composition of makes standard Gibbs energy of biosynthesis of phages different from that of their host cell. The more negative Gibbs energy of biosynthesis allows viruses to hijack their host cell metabolism [123, 130, 131]. Thus, biothermodynamic analysis reveals how the chemical structure of phages ultimately allows them to multiply inside their host cells, due to more negative Gibbs energy of biosynthesis.

Standard thermodynamic properties of biosynthesis of the T4 phage, Lambda phage, and *E. coli* live matter are given in tab. 6. For all three organisms, standard enthalpies of biosynthesis are slightly negative, meaning that the biosynthesis process is slightly exothermic. Standard entropies of biosynthesis are positive, due to the increase in the total number of particles during the process. On the product side of the biosynthesis reactions, the total number of particles is between 1.7 and 2, while on the reactant side it is between 0.6 and 0.8, tab. 5. The large number of particles on the product side is due to the released water molecules and synthetized biomass. Thus, the total number of particles increases during the process, leading to positive entropy change. On the other hand, all standard Gibbs energies of biosynthesis are

slightly negative, between –17.6 and –36.1 kJ/C-mol, tab. 6. This means that the biosynthesis process itself is a spontaneous process on the M9 medium. However, the standard Gibbs energies of biosynthesis are only slightly negative, meaning that the process would be very slow, which is inadequate for living organisms [6]. Thus, additional negative Gibbs energy must be made available by the catabolism to make the process proceed at the required rate [6].

Table 6 also shows that standard Gibbs energies of biosynthesis of the T4 and Lambda bacteriophages are between 1.5 and 2 times more negative than those of their host *E. coli*. The more negative Gibbs energy of biosynthesis allows the viruses to hijack the host cell metabolism [26, 108]. The negative Gibbs energy of biosynthesis represents the thermodynamic driving force for biosynthesis for both bacteria [4, 6] and viruses [26, 108]. Gibbs energy of biosynthesis is proportional to the biosynthesis rate, the rate of formation of new live matter [132]. Thus, due to their more negative Gibbs energy of biosynthesis, the T4 and Lambda phages are able to hijack the host cell metabolism.

Standard Gibbs energies of biosynthesis are defined under standard conditions: standard temperature of 298.15 K (25 °C) and pressure 1 bar, as well as unit activities of all reactants and products [118, 119]. Thus, using standard Gibbs energies of biosynthesis implies assuming that activities of all substances in the biosynthesis reaction do not greatly influence Gibbs energy of biosynthesis [6, 124, 128]. This is true in case of large absolute values of Gibbs energy [6, 124, 128]. However, $\Delta_{bs}G^0$ for the analyzed organisms on the M9 medium is not highly negative. Thus, it is interesting to see the influence of activities, which are given in tab. 7.

Table 7 gives standard Gibbs energies of biosynthesis with different corrections for activities of reactants and products. Approximating $\Delta_{bs}G$ with its standard value $\Delta_{bs}G^0$ is the least accurate method. A more accurate approach is to approximate activities of substances with their concentrations, giving the $\Delta_{bs}G_C$ values. Even more accurate is to include the activity coefficients of ions, which interact the strongest, using the extended Debye-Huckel equation. This results in the $\Delta_{bs}G_{DH}$ values. Finally, the most accurate solution is to include the activity of uncharged species, like glucose, giving the $\Delta_{bs}G$ values. The data in tab. 7 reveals that, regardless of the method used, the ratio of Gibbs energies of T4 and Lambda phages to that of their host cells remains similar. The bacteriophages always have a more negative Gibbs energy of biosynthesis than their host *E. coli*. However, since the absolute values of $\Delta_{bs}G$ are relatively small for all the analyzed organisms, it is good to include the activity corrections. For example, $\Delta_{bs}G^0$ of the Lambda phage is –36.1 kJ/C-mol, while after the activity correction, it becomes $\Delta_{bs}G$ = –29.54 kJ/C-mol. The activity corrections for the analyzed microorganisms are between 22 and 25% of the final corrected $\Delta_{bs}G$ value.

Catabolism

The Gibbs energies of biosynthesis of the analyzed microorganisms do not provide a sufficient driving force for their metabolism to proceed at required rates [4, 6, 133, 134]. Thus, additional driving force must be provided by the catabolism [4, 6, 133, 134]. The catabolism degrades the substrate molecules into simple products, releasing free energy to drive the metabolism [4, 6, 69]. Catabolism of the analyzed organisms was described using a catabolic-half reaction. Since all the analyzed organisms are growing on the same medium, their catabolic half-reactions are identical. The stoichiometry and thermodynamic properties of the catabolic half-reactions are given in tab. 8. The stoichiometry of the catabolic half-reaction is that of aerobic glucose catabolism. Standard enthalpy of catabolism $\Delta_{cat}H^0$ = –2802.74 kJ/C-mol is much more negative than that of biosynthesis, meaning that catabolism is highly exothermic. The highly negative $\Delta_{cat}H^0$ results in a high thermodynamic driving force for the catabolic half-

reaction: standard Gibbs energy of catabolism is $\Delta_{cat}G^0$ = –2871.74 kJ/C-mol. Due to the large absolute value of $\Delta_{cat}G^0$, the activity correction is only minor, changing it to $\Delta_{cat}G$ = –2861.93 kJ/C-mol. The highly negative Gibbs energy of catabolism provides the thermodynamic driving force that allows both bacteriophages and their host cells to multiply at required rates.

Driving force of growth and biomass yield

Table 9 shows standard Gibbs energies of growth (metabolism), $\Delta_{met}G^0$, and biomass yields, Y, for the analyzed organisms. Standard Gibbs energy of growth depends on the degree of reduction of and the number of carbon atoms in the substrate [124, 125]. Since all the analyzed microorganisms are growing on the same M9 medium with glucose, the substrate is the same for all – glucose. Glucose, $C_6H_{12}O_6$, has a degree of reduction of 4 and contains 6 carbon atoms. Substituting these values into eqs. (4) and (5) gives the driving forces of growth, $\Delta_{met}G^0$, for the analyzed microorganisms, which are shown in tab. 9. For all the analyzed organisms, $\Delta_{met}G^0$ are the same, since all are using the same substrate. The $\Delta_{met}G^0$ values were calculated using three methods: eq. (4) by Heijnen and van Dijken [125], eq. (5) by Liu *et al*. [126], and the average value for most microorganisms [6].

The $\Delta_{met}G^0$ value given eq. (5) is –409.7 kJ/C-mol, which is similar to the value for most microorganisms, –500 kJ/C-mol. On the other hand, the value given by eq. (4), –236.1 kJ/C-mol, is less negative. Both the bacteriophages and *E. coli* have identical $\Delta_{met}G^0$ values, since $\Delta_{met}G^0$ depends only on the substrate, which is identical for both.

Even though, the $\Delta_{met}G^0$ is identical for the phages and bacteria, standard Gibbs energies of biosynthesis are significantly different, $\Delta_{bs}G^0$. Thus, it seems that $\Delta_{bs}G^0$ decides which organism will win the competition for the metabolic machinery. The organism that controls the metabolic machinery is better adapted to the environment and has an advantage. Even though the difference in Gibbs energies of biosynthesis is double, one should have in mind that the values of $\Delta_{bs}G^0$ themselves are not very great. Thus, numerically, this advantage is such that it enables the multiplication of the bacteriophages, but does not endanger the survival of the bacteria, enabling both species to survive the competitive conditions.

Bacteria and phages are known to coexist in many ecosystems, where they interact [135, 136]. A dynamic equilibrium exists between the bacteria and phages [135, 136]. Thus, even though phages infect bacteria, they are not able to completely dominate the ecosystem and make the competition process irreversible. In the ecosystem, both the bacteria and the phages coexist. This observation can be explained by the small difference in Gibbs energies of biosynthesis of the bacteria and phages. On the other hand, Gibbs energies of metabolism are similar and explain the existence of both kinds of microorganisms. Figure 1 shows Gibbs energies of biosynthesis and metabolism of the analyzed organisms. From fig. 1 we see that Gibbs energies of metabolism are similar for all the analyzed organisms. On the other hand, Gibbs energies of biosynthesis of the phages two times more negative than that of *E. coli*.

Entire metabolism

Gibbs energies of metabolism were used to find biomass yields, tab. 9. The biomass yields were found for all three models. The values given by the model by Liu *et al.* [126] were chosen for further analysis, since the model has a better theoretical foundation and gives results more similar to that of most microorganisms (–500 kJ/C-mol) [6]. Biomass yields were used to find stoichiometric coefficients for the entire metabolism, which are presented in tab. 10, as well as standard enthalpy and entropy of metabolism (growth), given in tab. 11. The data from tabs. 10 and 11 were combined to find the influence of activities on Gibbs energy of metabolism, given in tab. 12.

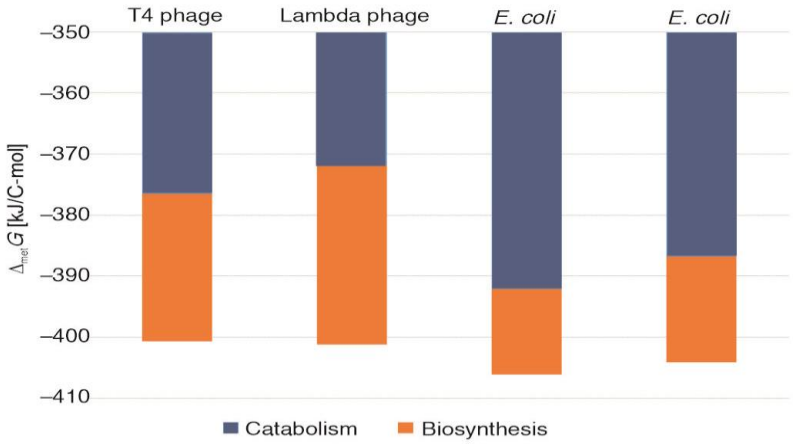

Figure 1. Gibbs energies of catabolism, biosynthesis and metabolism of T4 and Lambda phages, and their host *E. coli*; the contributions of Gibbs energies of catabolism ($\Delta_{cat}G$/Y, blue) and biosynthesis ($\Delta_{bs}G$, orange) to the total Gibbs energy of metabolism. Gibbs energies of metabolism are very similar for all the analyzed organisms, however, the values of $\Delta_{bs}G$ of the phages are two times larger than that of *E. coli*. *Note: for easier presentation, the graph begins at –350, not at 0*

Gibbs energy of metabolism has a relatively large value for the analyzed microorganisms, between –402 and –405 kJ/C-mol. Thus, the influence of activities of nutrients and metabolic products is relatively small. This is in accordance with the predictions of von Stockar *et al*. [128]. Therefore, it is good to take activities into account when discussing processes with a relatively low driving force, such as biosynthesis on the M9 medium. However, activities are less important when considering processes with a greater driving force, like entire metabolism and catabolism.

Conclusions

The driving forces for metabolism of bacteriophages and bacteria are identical - Gibbs energy. The driving force is shared between different classes of organisms, since they all evolved from the same last universal common ancestor (LUCA). The only differences is that the organisms took different pathways through evolution: bacteria evolved towards greater complexity, while phages evolved towards simplicity. However, bacteria and phages differ in the magnitude of Gibbs energy of biosynthesis. The phages have a more negative Gibbs energy of biosynthesis than bacteria, enabling them to hijack their metabolism. The absolute difference between Gibbs energies of biosynthesis of phages and bacteria is not very great. This allows both phages and bacteria to coexist in ecosystems.

Since the magnitudes of Gibbs energies of biosynthesis on M9 medium are not very great, the activities of nutrients and metabolic products change their values by between 22% and 25%. Thus, it is good to take activities into account when discussing biosynthesis on the M9 medium. On the other hand, Gibbs energies of catabolism and metabolism as a whole are much greater and activities make only a minor correction.

Nomenclature

(*Bio*) – live matter
A – constant in the extended Debye-Huckel equation
a_i – activity of substance i
$a_{ion,i}$ – effective diameter of the hydrated ion I [cm]

B – constant in the extended Debye-Huckel equation [Å^{-1}]
C_i – molarity of substance i [mol/dm^3]
I – ionic force of the solution
$n_{C,i}$ – number of C atoms in the formula of substance i
$n_{C,s}$ – number of carbon atoms in the substrate molecule
$n_{H,i}$ – number of H atoms in the formula of substance i
$n_{O,i}$ – number of O atoms in the formula of substance i
Q – reaction quotient
Q_C – activity reaction quotient
Q_c – concentration reaction quotient
Q_{DH} – Debye-Huckel reaction quotient
R – universal gas constant [Jmol^{-1}K^{-1}]
$S_m^0(i)$ – standard molar entropy of substance i [Jmol^{-1}K^{-1}]
T – temperature [K]
Y – biomass yield
z_i – charge of species i
$\Delta_{bs}G$ – Gibbs energy of biosynthesis [kJmol^{-1}]
$\Delta_{bs}G^0$ – standard Gibbs energy of biosynthesis [kJmol^{-1}]
$\Delta_{bs}G_C$ – Gibbs energy of biosynthesis with corrections for concentrations [kJmol^{-1}]
$\Delta_{bs}G_{DH}$ – Gibbs energy of biosynthesis with Debye-Huckel activity corrections [kJmol^{-1}]
$\Delta_{bs}H^0$ – standard enthalpy of biosynthesis [kJmol^{-1}]
$\Delta_{bs}S^0$ – standard entropy of biosynthesis [Jmol^{-1}K^{-1}]
$\Delta_{cat}G$ – Gibbs energy of catabolism [kJmol^{-1}]
$\Delta_{cat}G^0$ – standard Gibbs energy of catabolism [kJmol^{-1}]
$\Delta_{cat}H^0$ – standard enthalpy of catabolism [kJmol^{-1}]
$\Delta_{cat}S^0$ – standard entropy of catabolism [Jmol^{-1}K^{-1}]
$\Delta_fG^0(i)$ – standard Gibbs energy of formation of substance i [kJmol^{-1}]
$\Delta_fH^0(i)$ – standard enthalpy of formation of substance i [kJmol^{-1}]
Δ_rG – reaction Gibbs energy [kJmol^{-11}]
Δ_rG_C – reaction Gibbs energy with corrections for concentrations [kJmol^{-1}]
Δ_rG_{DH} – reaction Gibbs energy with Debye-Huckel activities [kJmol^{-11}]
$\Delta_{met}G$ – Gibbs energy of growth (metabolism) [kJmol^{-1}]
$\Delta_{met}G^0$ – standard Gibbs energy of growth (metabolism) [kJmol^{-1}]
$\Delta_{met}H^0$ – standard enthalpy of growth (metabolism) [kJmol^{-1}]
$\Delta_{met}S^0$ – standard entropy of growth (metabolism) [Jmol^{-1}K^{-1}]
Δ_rG^0 – standard reaction Gibbs energy [kJmol^{-1}]
Δ_rG^{0*} – reaction Gibbs energy with glucose activity correction [kJmol^{-1}]
Δ_rH^0 – standard reaction enthalpy [kJmol^{-1}]
Δ_rS^0 – standard reaction entropy [Jmol^{-1}K^{-1}]

Greek simbols

γ_i – activity coefficient of substance i
Γ_i – generalized degree of reduction of organic substance i
Γ_s – degree of reduction of the substrate molecules
ν_i – stoichiometric coefficient of substance i
$\nu_{i,bs}$ – stoichiometric coefficient of substance i in the biosynthesis half-reaction
$\nu_{i,cat}$ – stoichiometric coefficients of substance i in the catabolic half-reaction
$\nu_{i,met}$ – stoichiometric coefficient of substance i in the total growth (metabolic) reaction

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