ELSEVIER

Biophysical Chemistry 111 (2004) 115–122

Biophysical Chemistry

www.elsevier.com/locate/bpc

Standard apparent reduction potentials of biochemical half reactions and thermodynamic data on the species involved

Robert A. Alberty*

Department of Chemistry, Massachusetts Institute of Technology, 77 Mass. Ave., Cambridge, MA 02139, USA

Received 4 May 2004; received in revised form 17 May 2004; accepted 17 May 2004
Available online 1 June 2004

Abstract

Standard apparent reduction potentials E'° of half reactions of enzyme-catalyzed reactions are useful because they provide a global view of the apparent equilibrium constants of redox reactions. A table of E'° at a specified pH shows at a glance whether a given half reaction will drive another half reaction or be driven by it. This table can be used to calculate apparent equilibrium constants. Standard Gibbs energies of formation of species in a half reaction can be used to calculate E'° values at pHs in the range 5–9 and ionic strengths in the range of 0–0.35 M. My previously published values of E'° values for 42 half reactions has been extended by 22 new E'° values in this paper. When $\Delta_f G^{\circ}$ and $\Delta_f H^{\circ}$ are both known for all the species in an enzyme-catalyzed reaction at 298.15 K, it is possible to calculate all the standard transformed thermodynamic properties of the reaction over a range of pHs, ionic strengths, and temperatures.

Keywords: Apparent equilibrium constants; Electromotive forces; Redox reactions; Reduction potentials; Transformed thermodynamic properties; Transformed gibbs energy

1. Introduction

When the standard Gibbs energies of formation $\Delta_f G^{\circ}$ are known for all of the species in a biochemical redox reaction, the standard apparent reduction potentials E'° of the half reactions that are involved can be calculated as a function of pH and ionic strength [1,2]. In the first of these articles, standard apparent reduction potentials at 298.15 K, pH 7, and ionic strengths I of 0, 0.10, and 0.25 M were given in Table 7 for 31 half reactions for which $\Delta_f G^{\circ}$ (298.15 K, $I=0$) values were known for all of the species involved. In the second article, the half reactions involved in the methane monooxygenase reaction and the nitrogenase reaction were emphasized, and E'° values were given for 11 additional half reactions.

The most efficient way to store thermodynamic information on enzyme-catalyzed reactions is by means of small matrices that give $\Delta_f G^{\circ}$ (298.15 K, $I=0$), $\Delta_f H^{\circ}$ (298.15 K, $I=0$), z, and N_H for each species of the reactant. The charge number is represented by z, and the number of hydrogen atoms in a species is represented by N_H. The basic thermodynamic data on species in 131 reactants in enzyme-catalyzed reactions are available in BasicBiochemData2 [3] that can be downloaded into a computer with Mathematica^R [4] installed. These tables of thermodynamic data and computer programs are also available in printed form [5]. The objective of the present article is to extend this database with respect to redox reactions and to show how standard transformed thermodynamic properties vary with changes in pH, ionic strength, and temperature. The database on species is extended here to include certain oxides of nitrogen and sulfur for which data are available in the NBS Tables [6]. Standard thermodynamic properties of new organic species have been obtained from the literature on enzyme-catalyzed reactions. Goldberg et al. [7] have surveyed the literature on experimental measurements of K' and $\Delta_f H'^{\circ}$ and published evaluated compilations of data on oxidoreductases that has been a source of references for the current and previous papers [1,2].

When only $\Delta_f G^{\circ}$ are known for all species in a redox reaction at 298.15 K, it is possible to calculate apparent equilibrium constants, changes in binding of hydrogen ions, standard apparent reduction potentials, and changes in binding of hydrogen ions in half reactions at desired pHs and ionic strengths. When $\Delta_f H^{\circ}$ of species are known in addition, it is possible to calculate the effects of temperature on these

---

* Tel.: +1-617-253-2456; fax: +1-617-253-7030.
E-mail address: alberty@mit.edu (R.A. Alberty).

0301-4622/$ - see front matter © 2004 Elsevier B.V. All rights reserved.
doi:10.1016/j.bpc.2004.05.003

properties and on $\Delta_r H'^\circ$ and $\Delta_r S'^\circ$; in addition, it is possible to calculate E'° and the standard transformed enthalpies $\Delta_r H'^\circ$ and standard transformed entropies $\Delta_r S'^\circ$ of half reactions at desired temperatures, pHs, and ionic strengths.

2. Theory

The basic equations relating the standard apparent reduction potentials E'° and the standard transformed Gibbs energies $\Delta_f G'^\circ$ of half reactions and reactions have been given in the previous papers [1,2] and are reviewed in Ref. [5]. For the current paper, it is necessary to review the calculation of standard transformed enthalpies $\Delta_r H'^\circ$ of reaction, standard transformed entropies of reaction $\Delta_r S'^\circ$, and changes in the binding of hydrogen ions $\Delta_r N_H$ in reactions and half reactions at specified pH. These properties can all be calculated from the standard transformed Gibbs energy of reaction $\Delta_r G'^\circ$, expressed as a function of temperature, pH, and ionic strength, by taking partial derivatives [8]. The Gibbs–Helmholtz equation yields the function of temperature, pH, and ionic strength for the standard transformed enthalpy of redox reactions or half reactions:

$$\Delta_r H'^\circ = -T^2 \left(\frac{\partial (\Delta_r G'^\circ / T)}{\partial T} \right)_{P,\mathrm{pH}} \tag{1}$$

The fundamental equation for the transformed Gibbs energy shows that

$$\Delta_r S'^\circ = -\left(\frac{\partial \Delta_r G'^\circ}{\partial T} \right)_{P,\mathrm{pH}} \tag{2}$$

The change in binding of hydrogen ions in a reaction or half reaction is given by

$$\Delta_r N_H = \frac{1}{RT\ln(10)} \left(\frac{\partial \Delta_r G'^\circ}{\partial \mathrm{pH}} \right)_{T,P} \tag{3}$$

The standard apparent reduction potential E'° is related to the change in the standard transformed Gibbs energy of reaction by

$$\Delta_r G'^\circ = -|\nu_c| F E'^\circ \tag{4}$$

where $|\nu_c|$ is the number of formal electrons transferred and F is the faraday (96,485 C mol^{-1}). Substituting this equation into Eqs. (1)–(3) yields

$$\Delta_r H'^\circ = |\nu_c| F T^2 \left(\frac{\partial (E'^\circ / T)}{\partial T} \right)_{P,\mathrm{pH}} \tag{5}$$

$$\Delta_r S'^\circ = |\nu_c| F \left(\frac{\partial E'^\circ}{\partial T} \right)_{P,\mathrm{pH}} \tag{6}$$

$$\Delta_r N_H = -\frac{|\nu_c| F}{RT\ln(10)} \left(\frac{\partial E'^\circ}{\partial \mathrm{pH}} \right)_{T,P} \tag{7}$$

3. Redox reactions involving nitrogen

Standard apparent reduction potentials were calculated earlier [2] for reactions involved in the nitrogenase reaction (EC 1.18.6.1). It is convenient to refer to enzyme-catalyzed reactions by their Enzyme Nomenclature number [9], although strictly speaking these numbers apply to enzymes, rather than reactions. The values of $\Delta_f G^\circ$ and $\Delta_f H^\circ$ of NO_3^-(aq), HNO_3(aq), NO_2^-(aq), HNO_2(aq), NO(g), and N_2O(g) are available at 298.15 K in the NBS Tables [6]. These values are summarized in Table 1 along with species properties obtained later in this article.

Functions of pH and ionic strength that yield the standard transformed Gibbs energies of formation $\Delta_f G'^\circ$ of these reactants at 298.15 K were derived using the Mathematica program calcdGmat [10]. The data in Table 1 and Basic-

Table 1
Standard thermodynamic data on species at 298.15 K and zero ionic strength

Species	$\Delta_f G^\circ$/kJ mol^{-1}	$\Delta_f H^\circ$/kJ mol^{-1}	z	N_H
NO_3^-	− 108.74	− 205.00	− 1	0
HNO_3	− 111.25	− 207.36	0	1
NO_2^-	− 32.20	− 104.60	− 1	0
HNO_2	− 50.60	− 119.20	0	1
NO(g)	86.55	90.25	0	0
N_2O(g)	104.20	82.05	0	0
S(s)	0	0	0	0
S^{2-}	85.80	33.10	− 2	0
HS^-	12.08	− 17.60	− 1	1
H_2S	− 27.83	− 39.70	0	2
SO_4^{2-}	− 744.53	− 909.27	− 2	0
HSO_4^-	− 755.91	− 887.34	− 1	1
SO_3^{2-}	− 486.50	− 635.50	− 2	0
HSO_3^-	− 527.78	− 626.22	− 1	1
Butanal	− 129.87	–	0	8
Dihydroxyacetone	− 451.00	–	0	6
Xylitol	− 784.09	–	0	12
D-sorbitol	− 946.36	–	0	14
L-iditol	− 944.73	–	0	14
D-mannitol 1-phosphate^{2-}	− 1793.58	–	− 2	13
D-mannitol 1-phosphate^{-}	− 1830.68	–	− 1	14
D-glucono-1,5-lactone 6-phosphate^{2-}	− 1746.08	–	− 2	9
D-glucono-1,5-lactone 6-phosphate^{-}	− 1782.73	–	− 1	10
Ribitol	− 774.80	–	0	12
D-glucono-1,5-lactone	− 903.50	–	0	10
L-sorbitol 6-phosphate^{2-}	− 1793.10	–	− 2	13
L-sorbitol 6-phosphate^{-}	− 1829.75	–	− 1	14

The convention of this table is that the species are in dilute aqueous solution unless they are gases (indicated by g) or solids (indicated by s).

BiochemData2 (3.5) make it possible to calculate the apparent equilibrium constants of the following enzyme-catalyzed reactions at 298.15 K and pHs in the range 5–9 and ionic strengths in the range 0–0.35 M:

EC 1.7.1.3 Nitrate + $NADP_{red}$ = nitrite + H_2O + $NADP_{ox}$ $\|\nu_c\| = 2$	Nitrate reductase (NADP)
EC 1.7.1.4 Nitrite + 3 $NADP_{red}$ = ammonia + 2 H_2O + 3 $NADP_{ox}$ $\|\nu_c\| = 6$	Nitrite reductase (NADP)
EC 1.7.2.1 Nitrite + cytochrome c_{red} = NO(g) + H_2O + cytochrome c_{ox} $\|\nu_c\| = 1$	Nitrite reductase (NO forming)
EC 1.7.7.2 Nitrate + 2 ferredoxin$_{red}$ = nitrite + H_2O + 2 ferredoxin$_{ox}$ $\|\nu_c\| = 2$	Ferredoxin nitrate reductase
EC 1.7.99.6 (1/2) $N_2O(g)$ + cytochrome c_{red} = (1/2) $N_2(aq)$+(1/2) H_2O + cytochrome c_{ox} $\|\nu_c\| = 1$	Nitrous oxide reductase
EC 1.7.99.7 NO(g) + cytochrome c_{red} = (1/2) $N_2O(g)$+(1/2) H_2O + cytochrome c_{ox} $\|\nu_c\| = 1$	Nitric oxide reductase

The values of K' for these six reactions have been calculated at 298.15 K and five pHs at 0.25 M ionic strength using the Mathematica program calckprime [10]. These values are given in Table 2.

In all cases, K' decreases or remains constant with increasing pH. The apparent equilibrium constants in this table are too large to determine directly, except for reaction 1.7.2.1. The only pK involved in a significant way is that of ammonia (pK = 9.25), which is involved in reaction 1.7.1.4. The values of K' for reactions 1.7.1.3, 1.7.99.6, and 1.7.99.7 drop off by a factor of 10 per pH unit. The K' for reactions 1.7.2.1 and 1.7.7.2 drop off by a factor of 100 per pH unit. For reaction 1.7.1.4, the drop off is by a factor of 10^5 per pH unit. The $\Delta_r N_H$ are readily calculated using the Mathematica program calc$N_H$rx [5].

Each of the six redox reactions involving nitrogen can be divided into two half reactions. The standard apparent reduction potentials of these half reactions can be calculated at desired pHs and ionic strengths using the Mathematica program calcappredpots [2]. These half reactions are listed

Table 2
Apparent equilibrium constants K' for reactions involving nitrogen at 298.15 K and 0.25 M ionic strength

EC	pH 5	pH 6	pH 7	pH 8	pH 9
1.7.1.3	3.69×10^{26}	3.66×10^{25}	3.66×10^{24}	3.66×10^{23}	3.66×10^{22}
1.7.1.4	2.13×10^{76}	2.15×10^{71}	2.16×10^{66}	2.27×10^{61}	3.35×10^{56}
1.7.2.1	5.44×10^{6}	5.48×10^{4}	549	5.49	0.0549
1.7.7.2	3.07×10^{31}	3.05×10^{29}	3.05×10^{27}	3.045×10^{25}	3.05×10^{23}
1.7.99.6	1.51×10^{21}	1.51×10^{20}	1.51×10^{19}	1.51×10^{18}	1.51×10^{17}
1.7.99.7	1.22×10^{18}	1.22×10^{17}	1.22×10^{16}	1.22×10^{15}	1.22×10^{14}

Table 3
Calculated standard apparent reduction potentials E'° in volts of biochemical half reactions at 298.15 K and 0.25 M ionic strength[a]

Half reaction	pH 5	pH 6	pH 7	pH 8	pH 9
(1/2)$N_2O(g)$ + e^- = (1/2)$N_2(aq)$+ (1/2)H_2O	1.4649	1.4058	1.3466	1.2875	1.2283
NO(g) + e^- = (1/2)$N_2O(g)$+ (1/2)H_2O	1.2820	1.2229	1.1637	1.1045	1.0454
$O_2(aq) + 4e^- = 2H_2O$	0.9675	0.9083	0.8491	0.7900	0.7308
$O_2(g) + 4e^- = 2H_2O$	0.9250	0.8658	0.8066	0.7475	0.6883
Nitrate + $2e^-$ = nitrite + H_2O	0.5284	0.4692	0.4100	0.3508	0.2917
Nitrite + e^- = NO(g) + H_2O	0.6106	0.4924	0.3741	0.2558	0.1375
Glucose + $2e^-$ = glucono-1,5-lactone	0.2399	0.2991	0.3582	0.4174	0.4766
$O_2(aq) + 2e^- = H_2O_2$	0.4754	0.4162	0.3570	0.2979	0.2387
Nitrite + $6e^-$ = ammonia + $2H_2O$	0.4952	0.4163	0.3375	0.2588	0.1816
Glucose 6-phosphate + $2e^-$ = glucono-1,5-lactone 6-phosphate	0.2116	0.2708	0.3300	0.3891	0.4483
Sorbitol + $2e^-$ = fructose	0.1443	0.2035	0.2627	0.3218	0.3810
Sorbitol 6-phosphate + $2e^-$ = fructose 6-phosphate	0.1331	0.1943	0.2549	0.3143	0.3734
Iditol + $2e^-$ = sorbose	0.1343	0.1935	0.2527	0.3118	0.3710
Mannitol 1-phosphate + $2e^-$ = fructose 1-phosphate	0.1285	0.1908	0.2522	0.3118	0.3710
Xylitol + $2e^-$ = xylulose	0.1076	0.1667	0.2259	0.2851	0.3442
Ribitol + $2e^-$ = ribulose	0.1028	0.1620	0.2211	0.2803	0.3395
Cytochrome c_{ox} + e^- = cytochrome c_{red}	0.2121	0.2121	0.2121	0.2121	0.2121
Butanol + $2e^-$ = 1-butanal	0.0867	0.1458	0.2050	0.2642	0.3233
Glycerol + $2e^-$ = dihydroxyacetone	0.0633	0.1225	0.1816	0.2408	0.2999
Sulfite + $4e^-$ = sulfur(s) + $3H_2O$	0.0937	0.0187	− 0.0614	− 0.1480	− 0.2365
Sulfite + $8e^-$ = hydrogen sulfide(aq) + $3H_2O$	0.0069	− 0.0449	− 0.0968	− 0.1487	− 0.2004
Sulfite + $6e^-$ = hydrogen sulfide(aq) + $3H_2O$	0.0093	− 0.0598	− 0.1291	− 0.1982	− 0.2672
Glutathione$_{ox}$ + $2e^-$ = 2 glutathione$_{red}$	− 0.1732	− 0.2320	− 0.2876	− 0.3254	− 0.3359
Thioredoxin$_{ox}$ + $2e^-$ = thioredoxin$_{red}$	− 0.1727	− 0.2317	− 0.2888	− 0.3318	− 0.3438
NAD_{ox} + $2e^-$ = NAD_{red}	− 0.2569	− 0.2865	− 0.3160	− 0.3456	− 0.3752
$NADP_{ox}$ + $2e^-$ = $NADP_{red}$	− 0.2574	− 0.2870	− 0.3166	0.3461	− 0.3757
Cystine + $2e^-$ = 2 cysteine	− 0.2465	− 0.3055	− 0.3628	− 0.4088	− 0.4266

(continued on next page)

Table 3 (*continued*)

Half reaction	pH 5	pH 6	pH 7	pH 8	pH 9
Ferredoxin$_{ox}$ + e^- = ferredoxin$_{red}$	-0.4030	-0.4030	-0.4030	-0.4030	-0.4030
$2e^- = H_2(g)$	-0.3042	-0.3635	-0.4225	-0.4817	-0.5409
Sulfate + $2e^-$ = sulfite + H_2O	-0.3627	-0.4492	-0.5256	-0.5891	-0.6488

[a] Since cytochrome *c*, ferredoxin, and thioredoxin are proteins, their standard apparent reduction potentials will depend on the particular protein.

in Table 3 in descending order of their standard apparent reduction potentials at pH 7.

The difference between Table 3 and earlier tables [11,12] of E'° values is that the values in Table 3 have all been calculated from species properties in the NBS tables [6] and species properties calculated from measurements of apparent equilibrium constants, as described later in this paper. When species properties are known, it is possible to calculate E'° at other pHs and ionic strengths. Tewari and Goldberg [13] have found E'° (298.15 K, pH 7, $I = 0.25$ M) $= -0.265$ V for glutathione$_{ox}$ + $2e^-$ = 2glutathione$_{red}$, but further research will have to be done to obtain more accurate values.

When writing biochemical reactions or half reactions at specified pH, it is necessary to use names for reactants that will be interpreted as sums of species when a reactant is made up of two or more species. The redox reactions and half reactions at specified pH do not balance hydrogen atoms. The understanding in the thermodynamics of enzyme-catalyzed reactions is that hydrogen ions are supplied at the specified pH, so that it is not necessary to write hydrogen ions on the left side of the next to last half reaction in Table 3. The number of electrons $|\nu_c|$ involved in a half reaction can be calculated as follows: (1) Oxidized and reduced species are written down for a chemical half reaction in such a way that the oxidized species are on the left and the numbers of all types of atoms except hydrogen are the same on the two sides of the equation. (2) The chemical equation for the half reaction is balanced with respect to hydrogen atoms by inserting hydrogen ions. (3) This chemical equation is balanced with respect to electric charges by inserting formal electrons. The same number of electrons is calculated when different protonated forms of species are used. To obtain the form of the half reaction to be used when the pH is specified, the hydrogen ions in the chemical half reaction are deleted and reactant names that represent sums of species are used.

Since Table 3 is arranged in descending order of E'° at pH 7, the oxidized form in a half reaction will be reduced at pH 7 by the reduced form in any half reaction below it in the table. The entries in Table 3 can be used to calculate the apparent equilibrium constants in Table 2 using the following equation:

$$K' = \exp[|\nu_c|F(E_R'^\circ - E_L'^\circ)/RT] \tag{8}$$

$E_R'^\circ$ is for the half reaction higher in the table, and $E_L'^\circ$ is for the half reaction lower in the table. The biochemical reaction for which K' is calculated is obtained by adding the reverse form of the half reaction that is lower in Table 3 after multiplying by a factor to make the numbers of formal electrons the same in the two half reactions.

The half reaction for the reduction of molecular oxygen to water is included in Table 3 to show that oxidized forms $N_2O(g)$ and $NO(g)$ can in principle produce gaseous molecular oxygen at pH 7. The apparent equilibrium constants for $2NO_2(g) = N_2(aq) + 2O_2(g)$ and $4NO(g) = 2N_2O(g) + O_2(g)$ can be calculated, but their rates are unknown. The other end of the range for biochemical half reactions at -0.4225 V for pH 7 shows that molecular hydrogen can be produced by half reactions with more negative standard apparent reduction potentials.

4. Redox reactions involving sulfur

The values of $\Delta_f G^\circ$ and $\Delta_f H^\circ$ at 298.15 K are available in the NBS Tables [6] for the sulfur-containing species in Table 1. The standard transformed thermodynamic properties of the following reactions can be calculated:

EC 1.8.1.2 Sulfite + 3 NADP$_{red}$ = hydrogen sulfide + 3 NADP$_{ox}$ + 3 H_2O	Sulfite reductase (NADP)
EC 1.8.1.6 2 L-cysteine + NAD$_{ox}$ = L-cystine + NAD$_{red}$	Cystine reductase
EC 1.8.1.9 Thioredoxin$_{red}$ + NADP$_{ox}$=thioredoxin$_{ox}$ + NADP$_{red}$	Thioredoxin-disulfide reductase
EC 1.8.2.1 Sulfite + 2 cytochrome c_{ox} + H_2O= sulfate + 2 cytochrome c_{red}	Sulfite dehydrogenase
EC 1.8.3.1 Sulfite + $O_2(aq)$ + H_2O=sulfate + H_2O_2	Sulfite oxidase
EC 1.8.3.3 2 Glutathione$_{red}$ + $O_2(aq)$= glutathione$_{ox}$ + H_2O_2	Glutathione oxidase
EC 1.8.7.1 Sulfite + 3 ferredoxin$_{red}$ = hydrogen sulfide + 3 ferredoxin$_{ox}$ + 3 H_2O	Sulfite reductase (ferredoxin)
EC 1.13.11.18 Sulfur + $O_2(aq)$ + H_2O = sulfite	Sulfur dioxygenase

The values of K' for these reactions at 298.15 K and 0.25 M ionic strength calculated using data in Table 1 and BasicBiochemData2 [3] are given in Table 4.

Note that reaction 1.8.7.1 changes direction at about pH 8. The standard apparent reduction potentials for the half reactions involving sulfur compounds are given in Table 3 as a function of pH. Studies of the thermodynamics of solution of $SO_2(g)$ in water and of aqueous sulfur dioxide solutions by Goldberg and Parker [14] have yielded Henry law constants that make it possible to calculate apparent

Table 4
Calculated values of apparent equilibrium constants K' for reactions involving sulfur at 298.15 K and 0.25 M ionic strength

EC	pH 5	pH 6	pH 7	pH 8	pH 9
1.8.1.2	1.11×10^{27}	1.09×10^{23}	1.04×10^{19}	1.01×10^{15}	1.01×10^{11}
1.8.1.6	0.445	4.38	38.2	137	54.4
1.8.1.9	6.89×10^{5}	6.79×10^{7}	5.82×10^{9}	1.65×10^{11}	4.21×10^{11}
1.8.2.1	2.69×10^{19}	2.26×10^{22}	8.69×10^{24}	1.21×10^{27}	1.26×10^{29}
1.8.3.1	3.99×10^{59}	3.35×10^{58}	1.29×10^{57}	1.80×10^{55}	1.87×10^{53}
1.8.3.3	8.46×10^{21}	8.21×10^{21}	6.21×10^{21}	1.18×10^{21}	2.56×10^{19}
1.8.7.1	2.36×10^{21}	2.32×10^{14}	2.21×10^{7}	2.15	2.14×10^{-7}
1.13.11.18	1.19×10^{59}	1.42×10^{60}	3.68×10^{61}	2.64×10^{63}	2.53×10^{65}

equilibrium constants written in terms of SO_2(g) rather than sulfite.

5. Calculation of species properties for organic reactants from experimental apparent equilibrium constants

When the standard Gibbs energies of formation of species are known at 298.15 K for all the reactants but one in an enzyme-catalyzed reaction, the measurement of K' for the reaction at a given pH and ionic strength makes it possible to calculate the standard transformed Gibbs energy of formation of the one reactant under the experimental conditions. To calculate species properties, a pK is required if the one reactant is made up of two species. The standard Gibbs energies of formation of the species of the reactant at 298.15 K and zero ionic strength can be calculated using calcGef1sp [3] and calcGef2sp [10]. The following reactions are used here to obtain the species data for 10 more reactants in this way. The biochemical redox reactions for which there are experimental K' values are as follows [7]:

EC 1.1.1.1 n-butanol + NAD_{ox} = 1-butanal + NAD_{red}	Alcohol dehydrogenase
EC 1.1.1.6 Glycerol + NAD_{ox} = dihydroxyacetone + NAD_{red}	Glycerol dehydrogenase
EC 1.1.1.10 Xylitol + $NADP_{ox}$ = L-xylulose + $NADP_{red}$	L-xylulose reductase
EC 1.1.1.14 D-sorbitol + NAD_{ox} = D-fructose + NAD_{red}	L-sorbitol dehydrogenase
EC 1.1.1.15 D-iditol + NAD_{ox} = D-sorbose + NAD_{red}	L-iditol 2-dehydrogenase
EC 1.1.1.17 D-mannitol 1-phosphate + NAD_{ox}= D-fructose 6-phosphate + NAD_{red}	D-mannitol-1-phosphate 5-dehydrogenase
EC 1.1.1.49 D-glucose 6-phosphate + NAD_{ox}= D-glucono-1,5-lactone 6-phosphate + NAD_{red}	Glucose-6-phosphate 1-dehydrogenase
EC 1.1.1.56 Ribitol + NAD_{ox}=D-ribulose + NAD_{red}	Ribitol 2-dehydrogenase
EC 1.1.1.119 D-glucose + NAD_{ox}= D-glucono-1,5-lactone + NAD_{red}	Glucose 1-dehydrogenase (NADP)
EC 1.1.1.140 D-sorbitol 6-phosphate + NAD_{ox}= D-fructose 6-phosphate + NAD_{red}	L-Sorbitol-6-phosphate 2-dehydrogenase

Table 5
Experimental apparent equilibrium constants used to calculate $\Delta_f G^\circ$(298.15 K, 0.25 M) of species of reactants

EC	Reactant	Expt. K'	pH	I/M	pK	References
1.1.1.1	butanal	1.8×10^{-3}	8.3	0.03		[7]
1.1.1.6	dihydroxyacetone	2.38×10^{-5}	7.0	0.15		[15]
1.1.1.10	xylitol	2.97×10^{-4}	7.0	0.05		[7]
1.1.1.14	D-sorbitol	0.454	8.6	0.10		[16]
1.1.1.15	L-iditol	4.23×10^{-3}	7.0	0.05		[7]
1.1.1.17	D-mannitol 1-phosphate	5×10^{-3}	7.0	0.10	6.50	[17]
1.1.1.49	D-glucono-1, 5-lactone 6-phosphate	1.5	7.0	0.10	6.42	[18]
1.1.1.56	ribitol	1.79×10^{-2}	8.6	0.10		[16]
1.1.1.119	D-glucono-1, 5-lactone	3.4	6.4	0.10		[19]
1.1.1.140	L-sorbitol 6-phosphate	0.234	8.66	0.05	6.42	[20]

The experimental data on these reactions are summarized in Table 5. The three pKs needed for these calculations have not been measured, but they have been estimated from similar phosphate esters. The species data calculated from the experimental data in Table 5 are given in Table 1. This information makes it possible to calculate the apparent equilibrium constants of these 10 reactions at 298.15 K, pHs in the range 5–9 and ionic strengths in the range 0–0.35 M. The apparent equilibrium constants at 298.15 K, five pHs, and ionic strength 0.25 M have been calculated and are shown in Table 6.

The E'° of the half reactions involved in these 10 reactions have been calculated and are given in Table 3.

6. Effect of temperature

When $\Delta_f G^\circ$ (298.15 K, $I=0$) and $\Delta_f H^\circ$ (298.15 K, $I=0$) are known for all the species in an enzyme-catalyzed reaction, $\Delta_r G^\circ$ can be expressed as a function of temperature, pH, and ionic strength in the range of temperatures where $\Delta_f H^\circ$ (T, $I=0$) is essentially constant. Eqs. (1)–(3) show that $\Delta_r H'^\circ$, $\Delta_r S'^\circ$, and $\Delta_r N_H$ can then be obtained as functions of temperature, pH, and ionic strength by simply taking partial derivatives. A Mathematica program calcthprops that can be used to calculate the functions for $\Delta_f G'^\circ$, $\Delta_f S'^\circ$, $\Delta_f H'^\circ$, $\Delta_f C'_P{}^\circ$. and N_H has been published [8]. In order to make the three tables in this section, this program has been modified to yield $\Delta_f G'^\circ$, $\Delta_f H'^\circ$, $\Delta_f S'^\circ$, and N_H. This modified program that derives the functions for these four properties from the species matrices for a biochemical reaction or half reaction is referred to as calcthprops2. The effect of temperature on a redox reaction is illustrated with the formate dehydrogenase reaction:

EC 1.2.1.2 formate + NAD_{ox} + H_2O = CO_2tot + NAD_{red}

Table 6
Calculated values of apparent equilibrium constants K' of reactions at 298.15 K and 0.25 M ionic strength

EC	pH 5	pH 6	pH 7	pH 8	pH 9
1.1.1.1	1.76×10^{-6}	1.76×10^{-5}	1.76×10^{-4}	1.76×10^{-3}	1.76×10^{-2}
1.1.1.6	2.85×10^{-7}	2.85×10^{-6}	2.85×10^{-5}	2.85×10^{-4}	2.27×10^{-3}
1.1.1.10	8.61×10^{-6}	8.61×10^{-5}	8.61×10^{-4}	8.61×10^{-3}	8.61×10^{-2}
1.1.1.14	1.57×10^{-4}	1.57×10^{-3}	1.57×10^{-2}	1.57×10^{-1}	1.57
1.1.1.15	7.20×10^{-5}	7.20×10^{-4}	7.20×10^{-3}	7.20×10^{-2}	7.20×10^{-1}
1.1.1.17	4.56×10^{-5}	4.56×10^{-4}	4.56×10^{-3}	4.56×10^{-2}	4.56×10^{-1}
1.1.1.49	2.95×10^{-2}	2.95×10^{-1}	2.95	29.5	295
1.1.1.56	6.18×10^{-6}	6.18×10^{-5}	6.18×10^{-4}	6.18×10^{-3}	6.18×10^{-2}
1.1.1.119	0.267	2.67	26.7	267	2670
1.1.1.140	6.51×10^{-5}	7.67×10^{-4}	8.55×10^{-3}	8.70×10^{-2}	0.872

CO_2tot represents the sum of H_2CO_3, HCO_3^-, CO_2^{2-}, and CO_2(aq). The species properties at 298.15 K and zero ionic strength are available several places [3,5]. These vectors of $\Delta_f G'^\circ$, $\Delta_f H'^\circ$, $\Delta_f S'^\circ$, and $\bar{N}_H$ derived with calcthprops2 can then be added or subtracted to obtain the four very complicated functions for the formate dehydrogenase reaction. The Mathematica assignment operation /.x – > can then be used to make tables of standard thermodynamic properties. Table 7 gives the standard transformed thermodynamic properties for the formate reductase reaction at three temperatures, five pHs, and ionic strength 0.25 M. This table shows that the apparent equilibrium constant increases with pH, and this is due to the decrease in the binding of hydrogen ions (or, in other words, production of hydrogen ions) under all of these conditions. As the pH is increased, $\Delta_r H'^\circ$ becomes less favorable for reaction, but $\Delta_r S'^\circ$ becomes more favorable for reaction. The rapid increase in $\Delta_r S'^\circ$ with pH is a consequence of the increase in production of hydrogen ions at the higher pHs.

The same calculations are made on the two half reactions of the formate dehydrogenase reaction. Table 8 gives the properties for the half reaction $NAD_{ox} + 2e^- = NAD_{red}$ at 0.25 M ionic strength. The contribution of this half reaction to the $\Delta_r H'^\circ$ of the formate dehydrogenase reaction is independent of pH because NAD does not have p*K*s in this pH range. In this half reaction, there is an increase of exactly one mole of hydrogen ions in the half reaction. The last three rows of the table show the dependencies of standard apparent reduction potentials on temperature and pH. The table shows that the standard apparent reduction potentials are determined by the changes in transformed entropy, rather than the changes in the transformed enthalpies.

Table 7
Standard transformed thermodynamic properties[a] for the reaction formate + NAD_{ox} + H_2O = CO_2tot + NAD_{red} at 0.25 M ionic strength

	T/K	pH 5	pH 6	pH 7	pH 8	pH 9
$\Delta_r G'^\circ$	273.15	– 14.49	– 15.44	– 18.80	– 23.74	– 29.12
	298.15	– 14.08	– 15.38	– 19.41	– 24.90	– 30.94
	313.15	– 13.86	– 15.39	– 19.81	– 25.63	– 32.10
	273.15	– 19.13	– 16.49	– 12.52	– 11.36	– 10.00
$\Delta_r H'^\circ$	298.15	– 18.72	– 15.54	– 11.79	– 10.73	– 8.48
	313.15	– 18.43	– 14.94	– 11.34	– 10.27	– 7.34
	273.15	– 16.975	– 3.857	23.002	45.353	70.029
$\Delta_r S'^\circ$	298.15	– 15.542	– 0.509	25.568	47.562	75.329
	313.15	– 14.605	1.444	27.052	49.056	79.067
	273.15	– 0.05	– 0.38	– 0.86	– 0.99	– 1.09
$\Delta_r N_H$	298.15	– 0.08	– 0.45	– 0.89	– 1.01	– 1.15
	313.15	– 0.09	– 0.49	– 0.91	– 1.01	– 1.19

[a] The transformed Gibbs energies and transformed enthalpies are given in kJ mol^{-1} and the transformed entropies are given in J K^{-1} mol^{-1}.

Table 9 gives the properties for the half reaction CO_2tot + $2e^-$ = formate + H_2O at 0.25 M ionic strength. Here, there are significant changes with pH because of the p*K* of CO_2tot (6.08 at 298.15 K and 0.25 M ionic strength). The standard apparent reduction potentials are determined by the changes in the transformed entropies, rather than the changes in the transformed enthalpies.

The formate dehydrogenase reaction is obtained by subtracting the second half reaction from the first, and the standard transformed thermodynamic properties for the formate dehydrogenase reaction in Table 7 are obtained on the same way. Thus, we see that the two half reaction simply pass on their thermodynamic properties to the formate dehydrogenase reaction. These calculations can be made for a number of other redox reactions for which $\Delta_f H^\circ$ values are known for all the species.

7. Discussion

This article is another example of the usefulness of tables of $\Delta_f G^\circ$ and $\Delta_f H^\circ$ of species in biochemical reactions. If only $\Delta_f G^\circ$ is known for all the species in a redox reaction, $\Delta_r G'^\circ$, K', $\Delta_r N_H$, and E'° can be calculated for the reaction and its half reactions in the range pH 5–9 and ionic strengths from 0 to 0.35 M. Since very complicated functions are involved, a computer with a mathematical application is really needed to calculate the thermodynamic properties of biochemical reactions, and Mathematica [4] is well suited to these calculations because of its facilities for making symbolic calculations,

Table 8
Standard transformed thermodynamic properties for the half reaction $NAD_{ox} + 2e^- = NAD_{red}$ at 0.25 M ionic strength

	T/K	pH 5	pH 6	pH 7	pH 8	pH 9
$\Delta_r G'^\circ$	273.15	42.80	48.02	53.25	58.48	63.71
	298.15	49.57	55.28	60.99	66.70	72.40
	313.15	53.63	59.62	65.62	71.61	77.61
$\Delta_r H'^\circ$	273.15	− 31.35	− 31.35	− 31.35	− 31.35	− 31.35
	298.15	− 31.12	− 31.12	− 31.12	− 31.12	− 31.12
	313.15	− 30.95	− 30.95	− 30.95	− 30.95	− 30.95
$\Delta_r S'^\circ$	273.15	− 271.447	− 290.592	− 309.737	− 328.882	− 348.027
	298.15	− 270.625	− 289.770	− 308.915	− 328.060	− 347.205
	313.15	− 270.083	− 289.228	− 308.373	− 327.517	− 346.662
$\Delta_r N_H$	273.15	1	1	1	1	1
	298.15	1	1	1	1	1
	313.15	1	1	1	1	1
E'°	273.15	− 0.2218	− 0.2489	− 0.2760	− 0.3031	− 0.3302
	298.15	− 0.2569	− 0.2865	− 0.3160	− 0.3456	− 0.3752
	313.15	− 0.2779	− 0.3090	− 0.3400	− 0.3711	− 0.4022

The transformed Gibbs energies and transformed enthalpies are given in kJ mol^{-1} and the transformed entropies are given in J K^{-1} mol^{-1}. The standard apparent reduction potentials are given in volts.

taking partial derivatives, and constructing tables and plots.

If in addition the $\Delta_f H^\circ$ are known for all the species in a redox reaction, $\Delta_r G'^\circ$, K', $\Delta_r H'^\circ$, $\Delta_r S'^\circ$, $\Delta_r N_H$, and E'° can be calculated for this reaction and its half reactions. For redox reactions, tables of standard apparent reduction potentials provide a more global view of the apparent equilibrium constants of enzyme-catalyzed reactions since half reactions high in the table drive half reactions lower in the table.

In this paper, the table of about 42 E'° values published earlier [1,2] has been extended by 22 half reactions by use of values for inorganic species from the NBS Tables and by use of experimental values of K' of 10 enzyme-catalyzed reactions. Thus, Table 3 can currently be extended to 64 different half reactions. In the future this table of E'° values can be further extended by use of experimentally determined K' values of redox reactions in the literature [7].

Apparent equilibrium constants at 298.15 K can be calculated from any pair of half reactions in the E'° table; specifically $R = N(N-1)/2$, where R is the number of independent reactions and N is the number of different half reactions. Table 3 contains 30 half reactions and so $R = 30 \times 29/2 = 435$. However, there are not enzymes for all of these reactions.

Acknowledgements

I am indebted to Dr. Robert N. Goldberg (NIST) for many helpful discussions. Grateful thanks to NIH for support of this research by award number 5-RO1-GM48358-09.

Table 9
Standard transformed thermodynamic properties for the half reaction $CO_2tot + 2e^- = formate + H_2O$ at 0.25 M ionic strength

	T/K	pH 5	pH 6	pH 7	pH 8	pH 9
$\Delta_r G'^\circ$	273.15	57.29	63.46	72.06	82.23	92.84
	298.15	63.66	70.66	80.40	91.60	103.34
	313.15	67.49	75.01	85.42	97.24	109.70
$\Delta_r H'^\circ$	273.15	− 12.22	− 14.86	− 18.83	− 20.00	− 21.35
	298.15	− 12.40	− 15.58	− 19.33	− 20.39	− 22.64
	313.15	− 12.52	− 16.01	− 19.62	− 20.68	− 23.61
$\Delta_r S'^\circ$	273.15	− 254.472	− 286.736	− 332.739	− 374.235	− 418.055
	298.15	− 255.084	− 289.262	− 334.483	− 375.622	− 422.534
	313.15	− 255.478	− 290.672	− 335.425	− 376.573	− 425.729
$\Delta_r N_H$	273.15	1.06	1.38	1.86	1.99	2.09
	298.15	1.07	1.45	1.89	2.01	2.15
	313.15	1.09	1.49	1.91	2.01	2.19
E'°	273.15	− 0.2969	− 0.3289	− 0.3734	− 0.4261	− 0.4811
	298.15	− 0.3299	− 0.3662	− 0.4166	− 0.4747	− 0.5355
	313.15	− 0.3497	− 0.3887	− 0.4427	− 0.5039	− 0.5685

The transformed Gibbs energies and transformed enthalpies are given in kJ mol^{-1} and the transformed entropies are given in J K^{-1} mol^{-1}. The standard apparent reduction potentials are given in volts.

References

[1] R.A. Alberty, Calculation of standard transformed formation properties of biochemical reactants and standard apparent reduction potentials of half reactions, Arch. Biochem. Biophys. 358 (1998) 25–39.

[2] R.A. Alberty, Standard apparent reduction potentials for biochemical half reactions as a function of pH and ionic strength, Arch. Biochem. Biophys. 389 (2001) 94–109.

[3] R.A. Alberty, BasicBiochemData2 (2003). http://library.wolfram.com/infocenter/MathSource/797.

[4] Wolfram Research, 100 World Trade Center, Champaign, IL 61820–7237.

[5] R.A. Alberty, Thermodynamics of Biochemical Reactions, Wiley, Hoboken, NJ, 2003.

[6] D.D. Wagman, W.H. Evans, V.B. Parker, R.H. Schumm, I. Hallow, S.M. Bailey, K.L. Churney, R.L. Nuttall, The NBS tables of chemical thermodynamic properties, J. Phys. Chem. Ref. Data 11 (Suppl. 2).

[7] R.N. Goldberg, Y.B. Tewari, D. Bell, K. Fasio, E. Anderson, Thermodynamics of enzyme catalyzed reactions: Part 1. Oxidoreductases, J. Phys. Chem. Ref. Data 22 (1993) 515–582.

[8] R.A. Alberty, Effect of temperature on the standard transformed thermodynamic properties of biochemical reactions with emphasis on Maxwell equations, J. Phys. Chem., B 107 (2003) 3631–3635.

[9] E.C. Webb, Enzyme Nomenclature, Academic Press, San Diego, 1992. Current version available at http://www.chem.qmw.ac.uk/iubmb/enzyme/.

[10] R.A. Alberty, Inverse Legendre transform in biochemical thermodynamics: applied to the last five reactions of glycolysis, J. Phys. Chem., B 106 (2002) 6594–6599.

[11] P.A. Loach, in: G.D. Fasman (Ed.), Handbook of Biochemistry and Molecular Biology, 3rd ed., Physical and Chemical Data, vol. 1, CRC Press, Cleveland, OH, 1976, pp. 122–130.

[12] I.H. Segel, Biochemical Calculations, 2nd ed., Wiley, Hoboken, NJ, 1976.

[13] Y.B. Tewari, R.N. Goldberg, Thermodynamics of the oxidation–reduction reaction {2 glutathione$_{\mathrm{red}}$(aq) + NADP$_{\mathrm{ox}}$(aq) = glutathione$_{\mathrm{ox}}$(aq) + NADP$_{\mathrm{red}}$(aq)}, J. Chem. Thermodyn. 35 (2003) 1361–1381.

[14] R.N. Goldberg, V. Parker, Thermodynamics of solution of SO_2(g) in water and of aqueous sulfur dioxide solutions, J. Res. Natl. Bur. Stand. 90 (1985) 341–358.

[15] W.G. McGregor, J. Phillips, C.H. Suelter, Purification and kinetic characterization of a monovalent cation-activated glycerol dehydrogenase from *Aerobacter aerogenes*, J. Biol. Chem. 249 (1974) 31–32.

[16] M. Chakravorty, L.A. Viega, M. Bacila, B.L. Horecker, Pentose metabolism in *Candida*: II. The diphosphopyridine nucleotide-specific polyol dehydrogenase of *Candida utilis*, J. Biol. Chem. 237 (1962) 1014.

[17] J.B. Wolff, N.O. Kaplan, D-Mannitol 1-phosphate dehydrogenase from *Escherichia coli*, J. Biol. Chem. 218 (1956) 849.

[18] L. Glaser, D.H. Brown, Purification and properties of D-glucose-6-phosphate dehydrogenase, J. Biol. Chem. 216 (1955) 67.

[19] C.K. Lee, W.J. Dobrogosz, Oxidative metabolism in *Pediococcus pentosaceus* III Glucose dehydrogenase system, J. Bacteriol. 90 (1965) 653.

[20] M. Hirai, Purification and characterization of sorbitol-6-phosphate dehydrogenase from lequat leaves, Plant Physiol. 67 (1981) 221.