Available online at www.sciencedirect.com

ScienceDirect

Geochimica et Cosmochimica Acta 195 (2016) 293–322

Geochimica et Cosmochimica Acta

www.elsevier.com/locate/gca

Geobiochemistry of metabolism: Standard state thermodynamic properties of the citric acid cycle

Peter A. Canovas III [a,b,*], Everett L. Shock [a,b,c]

[a] *Group Exploring Organic Processes in Geochemistry (GEOPIG), Arizona State University, Tempe, AZ 85257, USA*
[b] *School of Earth and Space Exploration (SESE), Tempe, AZ 85257, USA*
[c] *School of Molecular Sciences, Arizona State University, Tempe, AZ 85257, USA*

Received 22 February 2016; accepted in revised form 20 August 2016; available online 29 August 2016

Abstract

Integrating microbial metabolism into geochemical modeling allows assessments of energy and mass transfer between the geosphere and the microbial biosphere. Energy and power supplies and demands can be assessed from analytical geochemical data given thermodynamic data for compounds involved in catabolism and anabolism. Results are reported here from a critique of the available standard state thermodynamic data for organic acids and acid anions involved in the citric acid cycle (also known as the tricarboxylic acid cycle or the Krebs cycle). The development of methods for estimating standard state data unavailable from experiments is described, together with methods to predict corresponding values at elevated temperatures and pressures using the revised Helgeson–Kirkham–Flowers (HKF) equation of state for aqueous species. Internal consistency is maintained with standard state thermodynamic data for organic and inorganic aqueous species commonly used in geochemical modeling efforts. Standard state data and revised-HKF parameters are used to predict equilibrium dissociation constants for the organic acids in the citric acid cycle, and to assess standard Gibbs energies of reactions for each step in the cycle at elevated temperatures and pressures. The results presented here can be used with analytical data from natural and experimental systems to assess the energy and power demands of microorganisms throughout the habitable ranges of pressure and temperature, and to assess the consequences of abiotic organic compound alteration processes at conditions of subsurface aquifers, sedimentary basins, hydrothermal systems, meteorite parent bodies, and ocean worlds throughout the solar system.

Keywords: Geobiochemistry; Citric acid cycle; Bioenergetics; Metabolism; Standard state thermodynamics; Geobiology

1. INTEGRATING BIOCHEMICAL CYCLES WITH GEOCHEMICAL PROCESSES

Microbial metabolisms take advantage of a huge diversity of geochemical energy sources. Electron donors (reductants) include hydrogen, methane and other light hydrocarbons, hydrogen sulfide, sulfur, thiosulfate, $S_4O_6^{2-}$, polysulfides, sulfide minerals, ferrous iron and ferrous minerals, carboxylic acids, hydroxy acids, alcohols, amino acids, and other organic compounds. Many organic compounds can also be fermented through disproportionation reactions. Electron acceptors (oxidants) range from oxygen to carbon dioxide and carbon monoxide, ferric iron, nitrate, nitrite, nitrous and nitric oxide, sulfate, thiosulfate, sulfite, and sulfur. Chemotrophic microorganisms can be aerobic or anaerobic, and autotrophic, heterotrophic, or mixotrophic. Considerable progress has been made by examining hundreds of reactions that couple electron donors and acceptors and generating frameworks for calculating energy and power supplies in diverse geochemical environments (McCollom and Shock, 1997; McCollom, 2000; Amend

---

* Corresponding author at: Group Exploring Organic Processes in Geochemistry (GEOPIG), School of Earth & Space Exploration, Arizona State University, Tempe, AZ 85257, USA.
E-mail address: pcanovas@asu.edu (P.A. Canovas).

http://dx.doi.org/10.1016/j.gca.2016.08.028
0016-7037/

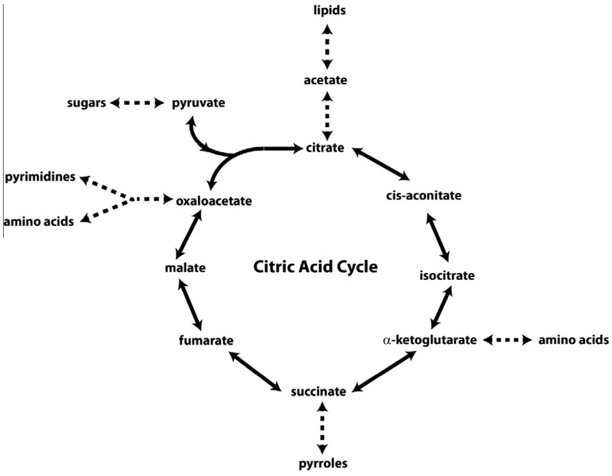

Fig. 1. A schematic depiction of the citric acid cycle. Clockwise corresponds to the forward (catabolic) direction, and counter-clockwise to the reverse (anabolic) direction, either of which is possible depending on the organism or the environment. Solid arrows indicate steps in the cycle. Dashed arrows indicate how the cycle connects to various processes integral to other metabolic pathways. The CAC is an integral part of overall metabolism and when run in reverse it is a powerful carbon fixation pathway connected to the production of lipids from citrate, sugars through pyruvate, amino acids through alpha-ketoglutarate and oxaloacetate, pyrroles through succinate, and pyrimidines through oxaloacetate.

and Shock, 2001; Amend et al., 2003, 2004, 2011, 2013; Shock, 2005; Rogers and Amend, 2006; McCollom, 2007; Windman et al., 2007; Skoog et al., 2007; Costa et al., 2009; Shock et al., 2010; Dodsworth et al., 2012; Frank et al., 2015). Meanwhile, less is known about the energy or power demands of microbes over wide ranges of temperature, pressure, and composition. With this in mind, we shift our focus inside the cell and onto the thermodynamics of the citric acid cycle (CAC), also known as the tricarboxylic acid cycle or the Krebs cycle, because it is central to many other biochemical processes.

Through the citric acid cycle, redox potential is captured from extracellular electron donors and acceptors that are far from equilibrium with one another and is used to provide the energy needed for biomolecule synthesis. Run in one direction, conventionally referred to as forward, the CAC is a major catabolic pathway in organisms for the aerobic oxidation of sugars, fatty acids, and amino acids, as shown in Fig. 1. Run in the opposite direction the reverse citric acid cycle (rCAC) is involved in anabolic CO_2 fixation into these and other organic metabolites. In the present study we developed an internally consistent set of standard state[1] thermodynamic properties and equation-of-state coefficients for compounds used in the citric acid cycle that are compatible with data for biomolecules that are linked to the CAC, as well as thermodynamic data for inorganic and organic aqueous species widely used in geochemical calculations. Implications of the thermodynamics of various steps in the CAC are discussed below, following an explanation of how internally consistent thermodynamic data were obtained.

2. STANDARD PARTIAL MOLAL THERMODYNAMIC PROPERTIES

Energy changes in geochemical reactions can be calculated by combining analytical data from natural environments with standard state thermodynamic properties of minerals, gases, organic compounds, and aqueous solutes. The approach often taken in theoretical geochemistry is to evaluate chemical affinities (**A**) using the relation

$$\mathbf{A} = 2.303RT\log(K/Q), \tag{1}$$

where R stands for the gas constant, T indicates temperature in Kelvin, Q represents the activity product for a given reaction, and K designates the equilibrium constant, which is related to the standard partial molal Gibbs energy of a reaction ($\Delta_r\bar{G}^o$) via

$$\Delta_r\bar{G}^o = -2.303RT\log K. \tag{2}$$

In turn, the standard partial molal Gibbs energy of a reaction is related to the standard partial molal Gibbs energies of the *ith* species in the reaction ($\bar{G}^o_i$) by

$$\Delta_r\bar{G}^o = \sum_i^r v_{i,r}\bar{G}^o_i \tag{3}$$

where $v_{i,r}$ represents the stoichiometric reaction coefficient of the *ith* species in the *rth* reaction, which is positive for products and negative for reactants. Values of $\bar{G}^o_i$ at elevated and pressures temperatures, $\bar{G}^o_{i,P,T}$, are calculated from

$$\bar{G}^o_{i,P,T} = \bar{G}^o_{i,P_r,T_r} - \bar{S}^o_{i,P_r,T_r}(T - T_r) + \int_{T_r}^{T}\bar{C}^o_{P,i}dT - T\int_{T_r}^{T}\bar{C}^o_{P,i}d\ln T + \int_{P_r}^{P}\bar{V}^o_i dP, \tag{4}$$

where $\bar{G}^o_{i,P_r,T_r}$ and $\bar{S}^o_{i,P_r,T_r}$ stand for the standard partial molal Gibbs energy and entropy of the *ith* species at the reference pressure of 1 bar and temperature of 298.15 K, and $\bar{V}^o_i$ and $\bar{C}^o_{P,i}$ represent the standard partial molal volume and isobaric heat capacity of the *ith* species, which are both functions of temperature and pressure. When the standard partial molal Gibbs energy of formation from the elements at the reference conditions, $\Delta\bar{G}^o_f$, is used in place of $\bar{G}^o_{i,P_r,T_r}$ then Eq. (4) represents the *apparent* standard partial molal Gibbs energy of formation of the *ith* chemical species at any temperature and pressure (Helgeson et al., 1981; Anderson, 2005). In addition, we make use of standard state enthalpies of reaction $\Delta_r\bar{H}^o$ that are related to standard state Gibbs energies of reaction by

$$\Delta_r\bar{G}^o = \Delta_r\bar{H}^o - T\Delta_r\bar{S}^o, \tag{5}$$

---

[1] The aqueous solution standard state adopted in this study is a hypothetical one molal solution referenced to infinite dilution at any temperature and pressure. The standard state for gases is the pure gas at any temperature and 1 bar, and that for liquid H_2O is the pure liquid at any temperature and pressure.