

Dirk Schulze-Makuch · Louis N. Irwin

## The prospect of alien life in exotic forms on other worlds

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**Abstract** The nature of life on Earth provides a singular example of carbon-based, water-borne, photosynthesis-driven biology. Within our understanding of chemistry and the physical laws governing the universe, however, lies the possibility that alien life could be based on different chemistries, solvents, and energy sources from the one example provided by Terran biology. In this paper, we review some of these possibilities. Silanes may be used as functional analogs to carbon molecules in environments very different from Earth; solvents other than water may be compatible for life-supporting processes, especially in cold environments, and a variety of energy sources may be utilized, some of which have no Terran analog. We provide a detailed discussion of two possible habitats for alien life which are generally not considered as such: the lower cloud level of the Venusian atmosphere and Titan's surface environment.

### Introduction

Is there other life out there? Would that life be similar to us or different, and how different could it be? This review tries to address that question, one of the oldest of human speculations. While at present we have no evidence that life exists anywhere other than on Earth, there are compelling reasons to assume that life exists pervasively throughout the universe (Schulze-Makuch and Irwin 2004). Life reflects the constraints of its environment and the resources available to it (Blum 1968). Thus, the structure of organisms and the na-

ture of their activities derive from the properties of matter and the course of evolution that gave rise to them—a point made by Henderson (1913) nearly a century ago and reiterated more recently by Gusev and Schulze-Makuch (2004). Where habitats similar to Earth have existed for a sufficient time, there is no reason to doubt that carbon-based, water-borne life may have evolved as it has in the one example with which we are familiar. Nonetheless, life cannot be excluded from habitats very different from those on Earth, as a different set of complex chemical interactions requiring different molecular components, solvent systems, and energy sources become possible at temperatures, pressures, and chemical compositions very different from those on Earth.

### A definition of life and its consequences

Any treatise about the possibility of alien forms of life has to begin with a definition of life. While there is no broadly accepted consensus on such a definition and an argument has even been made that defining life in the absence of a comprehensive theory and more cosmic set of examples may not be feasible (Cleland and Chyba 2002), guidance is needed nevertheless, if we hope to detect alien life in the solar system and beyond. Thus, given our current state of knowledge, we propose the following criteria for life: it is a complex system (1) composed of bounded compartments in thermodynamic disequilibrium with the external environment, (2) capable of transforming energy and the surroundings to maintain a low-entropy state, and (3) persisting through the encoding and transmission of information for self-replication. This preliminary definition, like many others with which it is comparable, (e.g., Dyson 1999; Koshland 2002; Monod 1971; Schrödinger 1944), places emphasis on a physical and thermodynamic distinction between the living system and its surroundings and on the central role of information. The definition of life given above could, in principle, apply to non-material phenomena; but as a practical matter, living systems—even exotic ones—are much more likely to be bounded, to interact precisely, and to

D. Schulze-Makuch (✉)  
Department of Geology,  
Washington State University,  
Pullman, WA 99164-2812, USA  
e-mail: dirksm@wsu.edu  
Tel.: +1-509-3351180  
Fax: +1-509-3357816

L. N. Irwin  
Department of Biological Sciences,  
University of Texas at El Paso,  
El Paso, TX, USA

encode information, as particles rather than as pure forces or energy. So we will restrict ourselves in this review to material forms of life with particular attention to the chemical components of the system, the solvent in which they interact, and the forms of energy that flow through the system to power its dynamic processes and maintain its low entropy state. Once those constraints have been delineated, we will then apply them to situations far different from those found on Earth, as an illustration of the diversity of forms that alien life could take.

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## Polymeric chemistry and macromolecules

Any life based on chemistry requires the presence of macromolecules and polymeric chemistry to interact with a versatile environment and a multitude of compounds. Macromolecules are required to address the very essential needs of how we define life in terms of compartmentalization, energy, and information, as noted above (Schulze-Makuch and Irwin 2004; Schulze-Makuch et al. 2002). Thus, a key feature of the functional structure of the chemistry of life can be seen to be highly specific non-repetitive macromolecules and their smaller components or interaction mediators (Bains 2004). Even if this life was based on molecular skeletons of atoms other than carbon, polymeric chemistry and macromolecules would still be required (e.g., Schulze-Makuch and Grinspoon 2005). During the origin of life phase, however, the first steps toward life may not have evolved biopolymers, but rather, non-covalent protocellular assemblies that were generated by the catalyzed recruitment of diverse amphiphilic and hydrophobic compounds. These could have constituted the first systems capable of information storage, inheritance, and selection (Segré and Lancet 2000), contrary to the more generally accepted prebiotic evolutionary scenario of the RNA world which is based on a very narrow subset of different chemical structures (Gesteland et al. 1999; Gilbert 1986; Sievers and von Kiedrowski 1994).

In this paper, we will examine how similar or different an alien biosphere could be from the Terran example and address the question of how different the biomolecules could be while still retaining their functional properties. We will also examine the possibility of more fundamental differences. The necessity of the three-biopolymer system (DNA–RNA–proteins) of information flow, for example, is questionable, as we know already from Terran organisms that some of them (viruses) do not require DNA, and presumably the first organisms in an RNA world functioned with neither proteins nor DNA. Then, we will consider whether some other element could replace carbon as a universal building block for life. Boron, nitrogen, silicon, phosphorus, and sulfur are other common non-metallic elements that are known to form heat-resistant polymers. In this context, we will discuss silicon, which has many properties similar to carbon and is the most promising substitute for carbon. It will be hard for any other element to match what we know about the complexity and versatility of carbon, but we have to keep in mind that (1) the research on

polymeric chemistry is carried out overwhelmingly under environmental conditions on Earth, and many polymers that are stable below the freezing point or above the boiling point of water are unknown; and (2) life may not, in principle, require any particular element to be as dominant as carbon is in Terran biochemistry. Chains composed of two or more different elements may work just as well (e.g., sequential units of B–N or Si–O; Schulze-Makuch and Irwin 2004). Proteins and nucleic acids already provide examples of structures that incorporate other elements into their backbones.

## Carbon chemistry different from Terran life

Life is based on complex chemistry, yet only a few of all the available elements participate in most life-supporting reactions on Earth: carbon, nitrogen, oxygen, hydrogen, phosphorus, and sulfur. Of these, the most characteristic element of biological systems is carbon. The great variety of structures formed from carbon, from chains and rings to three-dimensional macromolecules, are mostly stable within a broad temperature range, particularly under the surface conditions on Earth. The versatility of carbon is further enhanced by its ability to form double and triple bonds, which alters the chemistry and geometry of the molecule and the temperature-dependent fluidity of carbon-based molecular aggregates. The ability of carbon to build an almost unlimited range of molecules can be attributed to various factors: (1) the stability of carbon macromolecules due to a carbon–carbon bond energy that is higher than that of any other non-metal and comparable to the strengths of carbon–hydrogen and carbon–oxygen bonds; (2) carbon’s mid-range value of electronegativity that promotes the formation of primarily covalent bonds; and (3) high activation energies for substitution and bond cleavage reactions due to the absence of lone pairs or empty valence orbitals, thus, enhancing the stability of hydrocarbons to water and oxygen (Pannell, personal communication).

The ability of carbon to form the backbone of long-chain polymers is central to its contribution to the chemical complexity of living systems. The hydrocarbons that make up the long chain and cyclic skeleton of lipids consist exclusively of covalent carbon–carbon bonds. However, the make-up of the lipids is directly related to the solvent with which they interact. For example, in order for an organism to interface with a non-polar solvent, the membrane molecules would have to have hydrophobic headgroups instead of hydrophilic headgroups (see below “[Life on the surface of Titan?](#)”).

The backbones of proteins are repeating units of two carbon and one nitrogen atoms. These chains are made possible because of similar bond strengths between a carbon–carbon bond and a carbon–nitrogen bond. Proteins are made up of amino acids connected by peptide bonds. In Terran proteins, only 20 standard amino acids are used, but experiments with unnatural amino acids using the natural ribosome to incorporate them into proteins have expanded the amino acid repertoire of proteins (Bain et al. 1989; Hohsaka and Masahiko 2002; Noren et al. 1989). Furthermore, the

functionality of proteins is based on their efficient catalytic properties, which is related to their folding capacity. The folding capacity requires dipolar repeating elements, so that the positive ends of one dipole can interact with the negative ends of another dipole, inducing the tertiary structure of the protein. If that functionality is preserved, as found in polyamides, sulfonamides, or phosphoesters, there appears to be no reason why alternative sets of polypeptide chains and amino acids should be excluded from hypothetical proteins in putative alien life forms (Benner et al. 2004).

The backbone of nucleic acids is formed by repeating units of three carbons, one oxygen, one phosphorus, and another oxygen. The repeating element of RNA and DNA is a monopole (a charge) rather than a dipole. The capacity for each of these backbones to support an almost infinitely variable sequence of reactive functional side groups, while maintaining the stability of the skeletal framework, provides living systems with an almost endless repertoire of stable but variable and interchangeable molecular forms (Schulze-Makuch and Irwin 2004). Given the myriad of possibilities, how did Terran life adopt the particular structural forms of RNA and DNA that have evolved? Did they provide the best code available, resulting from a frozen accident in the process of the evolution of a prebiotic system, based perhaps on selective pressures that no longer exist (for a discussion see Gusev and Schulze-Makuch 2005)? First, the choice of the code is dependent on the solvent, and the solubility of DNA in water is beneficial to its stability (Westheimer 1987). However, even with water as a solvent, a variety of possibilities exist. Benner et al. (2004) pointed out the importance of phosphate linkages to molecular recognition in DNA and offered the following three hypotheses: (1) the repeating charges in the backbone force inter-strand interactions away from the backbone, causing the strands to contact at the edge of the heterocycles (without the polyanionic backbone, inter-strand contact could be anywhere (Steinbeck and Richert 1998)), (2) the repeating charges (monopoles) in the backbone keep DNA strands from folding, and (3) the repeating backbone charges allow DNA to support Darwinian evolution by generating inexact copies, with the inexactness itself being replicable. They further point out that the polyanionic backbones dominate the physical properties of DNA, and that replacing a nucleobase by another would thus, have only a second-order impact on the physical behavior of the molecule. Thus, different types of nucleotides can be expected in alien life. Miller et al. (1981) and Reddy and Bruise (2003) suggested that a polycationic backbone may work as well as a polyanionic backbone. Thus, the biochemistry of a genetic code in an alien organism may be quite different even though the same solvent (water) is used.

Energetically favorable redox reactions are the basis for Terran metabolism. Carbon can be converted fairly easily between its fully reduced state (methane, CH<sub>4</sub>) with a valence of -4 and its fully oxidized state (carbon dioxide, CO<sub>2</sub>) with a valence of +4, which makes it a favorable element for use in metabolism. It is a considerable advantage that both products are gases under a fairly broad temperature range including Earth environmental conditions. These compounds are the end products of metabolic

activity—carbon dioxide for aerobic respiration and methane for methanogenesis—and, because they are gases, they can be disposed of readily as metabolic end products. The nature of the specific metabolic strategy employed depends largely upon the environmental constraints that affect an organism. For instance, anaerobic bacteria use metabolic pathways that do not depend on oxygen. Organisms usually adapt to environmental conditions by evolving in a manner that allows them to utilize the raw materials available. This raw material can be any compound that can function as an electron acceptor or donor, in principle. For example, organisms are known that use As, Se, or U (Lovley et al. 1991; Stolz and Oremland 1999). The same could be expected for any alien organism. However, in modern Terran metabolism, the electrons generally move, directly or indirectly, to an oxygen atom that begins the reaction doubly bonded to a carbon atom (Benner et al. 2004). In an alien biochemistry with, for example, ammonia as solvent, the nitrogen doubly bonded to carbon could support the same function. Metabolic pathways for Earth organisms are very complex. The internal steps of the multi-step metabolic pathways are usually only slightly energetically downhill but require the last step to be energetically large in order to pull the reaction to completion (Voet and Voet 2004). Benner et al. (2004) suggested that this feature may be universal for metabolic pathways for any organism including alien ones because this procedure harvests most economically a surrounding chemical disequilibrium without wasting large amounts of free energy.

### Silicon chemistry

A biochemistry based on polymers of any atom other than carbon typically receives little serious scientific attention, other than in science fiction. However, this notion is mostly based on the vast improbability of silicon-based life under Earth environmental conditions. Recent work focusing on possible life in non-polar solvents and cryogenic environments has brought this common assertion into question. It has become increasingly clear that silanes can take part in a versatile chemistry under conditions not exposed to Earth's atmosphere. Silanes and their low molecular weight derivatives can form flexible, macromolecular assemblies in the form of sheets, strings, tubes, and other shapes, similar to those formed by lipid bilayers in carbon biochemistry (Lickiss et al. 1991, 1993; Unno et al. 2000). Silicon does occur in variable valences (4, 5, and 6) and forms stable covalent bonds with itself, carbon, germanium, nitrogen, phosphorus, oxygen, sulfur, halogens, and many metals (Muller et al. 1998; Walsh 1981). Some of the compounds that silicon can form include (1) stable tetra-, penta-, and hexa-coordinate compounds with N, C, and O bonds, with or without overall charge on the molecule (Bains 2004), (2) branched and unbranched chains and ring systems such as in cyclohexasilane (Schulze-Makuch and Irwin 2004), (3) highly polarized systems where rapid, reversible chemistry occurs (Sharma and Pannell 1995), (4) cage systems such as silsesquioxanes that provide for complex core

structures, which may be surrounded with other groups that are oriented precisely in space (Feher 2000; Harrison 1997) and may protect the core from degradation. Benner et al. (2004) pointed out that oligosilanes having up to 26 consecutive Si–Si bonds are known that can be (1) chiral, (2) support a variety of functionalized and non-functionalized side chains, (3) have alkyl side chains that are generally soluble in non-polar solvents, (4) carry carboxylic acid groups that are soluble in water, and (5) self-aggregate into amphiphilic structures in water, creating vesicles and micelles. Silanes cannot form a pi-conjugated system; hence, they do not form aromatic compounds. However, silicon can form sigma-conjugated polysilanes (Maxka et al. 1991) with electronic properties similar to those of the carbon-containing pi-conjugated system (Benner et al. 2004). In considering biochemical alternatives to carbon, the key is to look for functional rather than structural analogues. Bains (2004) demonstrated this principle by elaborating on the possibility of photosynthesis in silicon biochemistry. He pointed out that silicon's  $sp^2$  structure is disfavored over carbon's  $sp^3$  structure in its ability to delocalize charge over a large structure. However,  $sp^3$  silicon can delocalize electrons via s-orbital overlap to a degree that carbon cannot, to the extent that polysilanes are semiconducting (Tokito and Okazaki 1998; West 2001). Electron delocalization could therefore, occur readily in appropriate silicon compounds, and light-activated electronic effects (the basis of Terran photosynthesis) could also occur (Fujino 1987). Dahn et al. (1993) pointed out that layered silanes synthesized from calcium silicide of the general formula  $(SiH)_n$  have a layered structure and are electroluminescent.

Environmental conditions may exist for which biochemistry based on silanes might be a realistic possibility. They are: (1) a reducing atmosphere with only minor amounts of oxygen to prevent oxidation, (2) scarcity of water to prevent conversion into non-reactive silicates, (3) temperatures far below the freezing point of water to enhance the stability of weakly bonded complex Si–Si compounds, (4) a compatible solvent such as methane, and (5) a limited availability of carbon due to a greater chemical versatility of carbon that may out-compete silicon as a basic building block. Restriction 5 is based not only on chemistry under Earth environmental conditions but also based on material found in the heads of comets, inside meteorites, within the nebulae, and among the interstellar matter of the universe (Fegley 1987; Gladstone et al. 1993; Hanon et al. 1996; Llorca 1998; Varela and Metrich 2000). While polymeric carbon compounds seem to be ubiquitous, polymeric silicon compounds are not. There are now 140 different chemical species detected in space, most of them carbon compounds and only very few are silicon compounds (as of July 2005, see "<http://www.astrochemistry.net>"). Nevertheless, there are some environments where the formation of silicon polymers should be feasible, and such environments should not be uncommon. Bains (2004) noted the protostellar disc W33A in which it appears that 1/250 to 1/40 of all infrared-visible silicon atoms have a hydrogen directly bonded to them. Examples of processes and

environments suitable for creating silanes include: (1) serpentinization reactions in a cryogenic environment, (2) meteorite impacts on cold planetary bodies, and (3) ice/silicate grains exposed to UV radiation and 1 MeV protons in a hydrogen atmosphere (Bains 2004).

Thus, a silicon-based biochemistry has to be considered feasible in selected types of environments. Even within the carbon-dominated chemistry on Earth, silicon plays a biological role. Carbon compounds interact with silicon in diatoms to build a distinct organism. Silaffins, a type of unique peptide, and long-chained polyamines are constituents of the biosilica of diatoms and can precipitate silica nanospheres (Kröger et al. 2002). The formation of the biosilica occurs within a membrane-bound compartment (silica deposition vehicle) that acts as a casting mold (Pickett-Heaps et al. 1990). The carbon compounds appear to act as a template for silicon structures in diatoms. Cairns-Smith (1982, 1985); Cairns-Smith and Hartman (1986) suggested that silicon compounds may have provided the first templates for carbon-based life, noting that the first carbon compounds may have received their initial handedness from clays or silicate minerals that also had handedness. Thus, even in a carbon-dominated biochemistry, silicon may play a significant role. Likewise, other atoms such as oxygen, nitrogen, sulfur, phosphorus, or boron may be utilized in a biochemical framework.

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### Alternative solvents for life—is water really necessary?

Life, as we know it, consists of chemical interactions that take place in the liquid state. This is so because macromolecules need to be physically stable, yet capable of structural flexibility and chemical interactivity, which in practice means that they are able to move with respect to each other but are kept in constant close proximity (Bains 2004; Schulze-Makuch and Irwin 2004). This can occur only in a liquid state. The functions of a good solvent include: (1) an environment that allows for the stability of some chemical bonds to maintain macromolecular structure, while (2) promoting the dissolution of other chemical bonds with sufficient ease to enable frequent chemical interchange and energy transformations from one molecular state to another; (3) the ability to dissolve many solutes while enabling some macromolecules to resist dissolution, thereby, providing boundaries, surfaces, interfaces, and stereochemical stability; (4) a density sufficient to maintain critical concentrations of reactants and constrain their dispersal; (5) a medium that provides both an upper and lower limit to the temperatures and pressures at which biochemical reactions operate, thereby, funneling the evolution of metabolic pathways into a narrower range optimized for multiple interactions; and (6) a buffer against environmental fluctuations (Schulze-Makuch and Irwin 2004). The solvent properties of water and some other potential solvents are shown in Table 1.

Water is usually portrayed as the universal solvent for life as we know it, because of various properties that make it a very good solvent for the environmental conditions of Earth. These include a high dipole moment, which allows

**Table 1** Selected physical properties of water and some other solvent candidates

Property	H <sub>2</sub> O	NH <sub>3</sub>	HCN	H <sub>2</sub> SO <sub>4</sub>	H <sub>2</sub> S	CH <sub>3</sub> OH	CH <sub>3</sub> NO	N <sub>2</sub> H <sub>4</sub>	CH <sub>4</sub>	C <sub>2</sub> H <sub>6</sub>	N <sub>2</sub>
Molecular weight	18.015	17.031	27.02	98.08	34.08	32.04	45.04	32.05	16.04	30.07	28.01
Density (g/ml)	0.997	0.696	0.684	1.831	1.393	0.793	1.13	1.004	0.426 (at -164°C)	0.572 (at -107°C)	0.85
Melting point (°C at 1 bar)	0.00	-77.73	-13.29	-75.5	-85.5	-94	3	1.6	-182	-172	-210
Boiling point (°C at 1 bar)	100.0	-33.33	26.0	-10.0	-59.6	65	211	113.5	-161.5	-89	-196
Range of liquid (°C at 1 bar)	100	44.4	39.3	65.5	25.9	159	208	111	20.5	83	14
Critical temperature (°C)	374	132	184	NA	100	240	NA	380	-82.6	32.3	-147
Critical pressure (bar)	215	111	54	NA	88	78	NA	14.2	45.4	47.8	33.3
Enthalpy of fusion (kJ/mol)	6.0	5.7	8.4	10.7	2.4	2.2	8.44	12.7	0.94	2.7	0.71
Enthalpy of vaporization (kJ/mol)	40.7	23.3	25.2	NA	18.7	40.5	60.1 (at 25°C)	40,900	8.2	14.7	5.56
Dielectric constant ( $\epsilon$ )	80.1	16.6	114.9	101	5.9	354 (13°C)	110	51.7	1.7	1.9	1.45
Viscosity (10 <sup>-3</sup> P)	9.6	2.7 (-34°C)	2.0 (20°C)	260	4.3 (61°C)	5.9	33	9.8	0.009 (20°C)	0.011 (20°C)	2.04
Dipole moment ( <i>D</i> )	1.85	1.47	2.99	2.7	0.98	1.6	3.4	1.9	0	0	0
Surface tension (10 <sup>-3</sup> J/m <sup>2</sup> )	71.99	19.8	18.1	NA	NA	22.1	57.03	66.39	NA	NA	10.53
Relative probability of occurrence <sup>a</sup>	(25°C)	0.25	(22°C)	0.42	1.31	NA	(25°C)	(25°C)	0.62	1.25	1.96

Data from CRC (2001), Dean (1992), Firsoff (1963), Merck Research Labs (1996), Moeller (1957), [http://www.trimen.pl/witek/ciecze/old\\_liquids.html](http://www.trimen.pl/witek/ciecze/old_liquids.html), <http://www.flexwareinc.com/gasprop.htm>, and [http://chemweb.ump.ac.za/chemistry/Physical\\_Data/Solvent\\_properties.htm](http://chemweb.ump.ac.za/chemistry/Physical_Data/Solvent_properties.htm)  
 NA not available

<sup>a</sup>Relative probability of liquids on bodies >1,000 km in diameter around any star in our stellar neighborhood (from Bains 2004)

liquid water molecules to stick to each other via hydrogen bonding, providing it with polymer-like properties. The hydrogen bonding also raises the freezing and boiling point of water to much higher temperatures than it would otherwise be. One of the most biologically favorable properties of water is its role as a thermal moderator. The reason is that (1) water's high heat capacity allows it to be available over a wide range of temperatures (from 0 to 100°C at 1 bar pressure), and (2) water is a good heat insulator. Furthermore, the surface tension of water is very high allowing organic macromolecules to concentrate in microscopic interfaces. Water acts as a climatic stabilizer. Associated with that property is its unusual capacity to decrease in density as it freezes below its maximum density at 4°C. Thus, the floating, insulating layer of lighter-water ice maintains a liquid environment for organisms beneath its surface despite the freezing ambient temperatures above. Water also provides stability against environmental fluctuations in a chemical sense. Water dissociates into a proton, which is usually attached to a water molecule as a hydronium ion, and an anion, providing acid and base characteristics, further increasing the solubility of compounds immersed in water. Drastic changes in pH, which could easily be detrimental to life, can be prevented by the presence of naturally and widespread occurring buffers such as carbonate and phosphate. One other important environmental advantage of water is that in an Earth-type environment it is self-shielding against ultraviolet radiation. Ultraviolet photons dissociate water molecules, releasing oxygen and hydrogen into the atmosphere. If that happens, some of the oxygen atoms will form ozone molecules, which are excellent absorbers for UV radiation, thus, shielding the planetary surface from this detrimental form of high energy (Schulze-Makuch and Irwin 2004). If alien life requires proteins, which are essential biomolecules to life on Earth, water is an ideal solvent. Proteins fold to a unique, globular conformation which tightly packs over 80% of the peptide groups and non-polar side chains. This marginally higher stability of proteins in the folded state, which allows both stability and solubility, would be very difficult to reproduce in alternative solvents (Pace et al. 2004). Finally and perhaps most importantly, water is abundant not only on Earth but also in the universe based on spectral analyses. Thus, water is likely a good solvent for other planets as well, though many properties that make water such a good solvent are directly linked to the environmental conditions existing on Earth.

There are also negative properties of water. Water is not a good solvent for molecules with non-polar covalent bonds such as those of hydrophobic organic molecules like lipids, which serve as the core of cellular membranes and proteins embedded in the membrane core. Many organic synthesis reactions involve dehydration which is inhibited in the presence of water, hence, requiring energetically expensive and elaborately catalyzed reactions. Water is very reactive and therefore, damaging to many biomolecules including high-energy phosphatidyl compounds that break down easily and many cellular macromolecules, particularly the nucleic acids (Feinberg and Shapiro 1980). DNA has to be

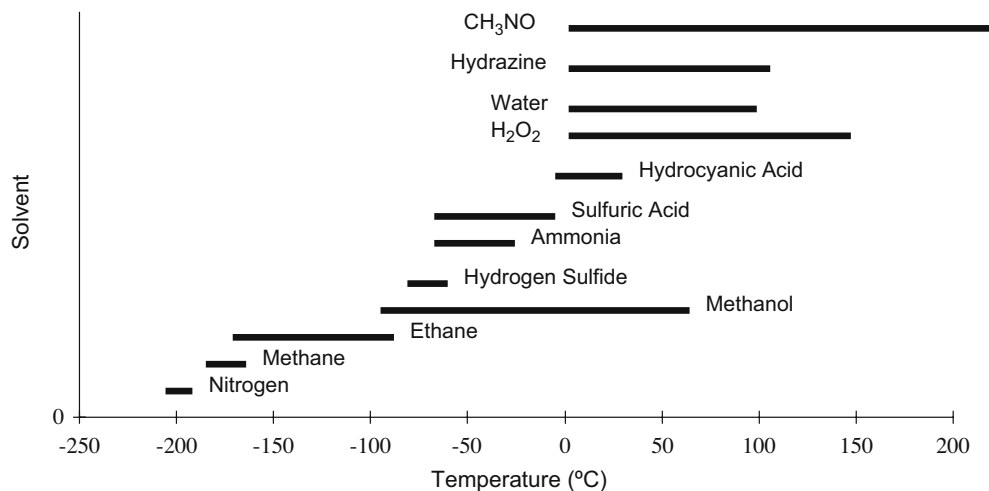
continuously repaired, and metabolic evolution has evolved generally under the limitations imposed by water on biochemical processes. The peculiar property of water having its highest density at 4°C also poses a disadvantage. Ice crystals pierce cellular membranes due to the expansion of water as a solid. This is the basis for frost damage to plants, for example. Benner et al. (2004) pointed out that many organic molecules that are parts of carbon-based metabolisms display problematic reactivity with water and elaborated on the difficulty with carbon dioxide cycling. At a recent symposium on the biological importance of water, Daniel et al. (2004) wondered about the biological functions of water at the molecular level and whether water is truly irreplaceable for the function of living systems.

### Ammonia

The molecular alternative to water, cited most often as a potential life-sustaining solvent, is ammonia. Ammonia has a structure comparable to water but a lower dipole moment and is thus less efficient at dissolving polarized compounds. However, it still dissolves many organic compounds. Ammonia's heat of vaporization and heat of fusion are lower than that of water. Thus, ammonia is not as good in moderating temperature as water but is still sufficient to have a stabilizing effect on climatic conditions. The surface tension of ammonia is only about one third that of water; thus, ammonia is not likely to concentrate organic macromolecules in microscopic interfaces as well as water. Ammonia dissociates into nitrogen and hydrogen and does not afford any protection from UV light when compared to the dissociation of water. Thus, any origin of life in ammonia would have to occur in some kind of protected environment. Pure ammonia, however, dissolves alkali metals without reaction, which is of biological relevance because alkali metals can act as catalysts. Also, salt solutions in liquid ammonia have usually a greater electrical conductivity than an aqueous solution of the same salt (Mee 1934). Ammonia is about four times less viscous than water, thus, dissolved particles and ions have a less difficult time to find and react with each other. Ammonia self-dissociates as water does but to a much lower degree; thus, acid-base reactions do occur in an ammonia system but to a much lower extent. Ammonia is certainly ubiquitous in the universe as it is found in the interstellar medium and in comets.

Ammonia has a smaller range in which it stays liquid, thus, life would have to adapt to a smaller temperature range to survive in such a solvent (Fig. 1). However, at higher pressure, it stays liquid within a wide temperature range (e.g., 196–371 K at 60 bar). As solid ammonia is denser than liquid ammonia, there is no mechanism to prevent liquid ammonia from completely freezing in ambient environments below its freezing temperature. This is no problem for microbial life, as microbes could have adapted to survive episodic or cyclic freezing in a dormant state (e.g., as spores). Furthermore, chemical reactions at colder temperatures would generally be expected to prog-

**Fig. 1** Temperature ranges for some solvent candidates to occur in the liquid state (at 1 bar)



ress at a slower pace, lengthening life spans, and therefore, the rate at which competition and adaptive radiation would drive evolution. Finally, liquid ammonia cannot co-exist with free oxygen, thus, aerobic metabolism would be inconsistent with ammonia as a solvent.

Biochemistry with ammonia as solvent would have to be different but not necessarily by much. Life on Earth exploits compounds containing the C=O carbonyl unit. In ammonia, C=O units would have to be replaced by C=N units, but given this adjustment, metabolism in liquid ammonia is easily conceivable (Benner et al. 2004). Haldane (1954) pointed out ammonia analogues to water and suggested the possibility of building proteins, nucleic acids, and polypeptides within a liquid ammonia solvent. Raulin et al. (1995) suggested that “ammono” analogues of Terran biomolecules in which oxygen atoms are replaced by NH groups might yield an equally viable pseudo-biochemistry. Firsoff (1963) went into some detail showing similarities of synthesis reactions in water-based, ammonia-based, and water–ammonia mixtures. An especially interesting example provided by him is the synthesis of proteins from amino acids through a peptide bond. While in a water system two glycine molecules combine with the release of water, in a water–ammonia mixture, the COOH group would be replaced with a CONH<sub>2</sub> group and ammonia would be released instead of water, while the peptide bond is preserved. A further parallel to water is that ammoniation reactions in ammonia as solvent are directly analogous to hydration reactions with water in which a salt and the solvent are produced. This leads to an important environmental consequence that minerals in a near-surface or subsurface “hydrosphere” would contain ammonia in their crystal structure just as Earth’s rocks contain water. Ammonia and water are definitely related solvents, as indicated by the fact that life sustaining organic macromolecules such as proteins, amino acids, and nucleic acids contain both OH and NH<sub>2</sub> functional groups in various combinations and proportions with which ammonia could easily interact. Aspinall et al. (2002) claimed that phosphate, critical for Terran life, can be replaced by ammono-

phosphate analogues that are stable in water and ammonia, can lose protons to hold multiple negative charges, and can form stable amide-like bonds with carbon molecules. Bains (2004) added that “electricity,” movement of electrons, rather than “proticity” would be a reasonable energy trans- action for ammonium supported life. Several enzymes of Terran organisms interestingly remain active down to temperatures of about –100°C (Bragger et al. 2000), well within the stability of an ammonia–water mixture.

Liquid ammonia could present an opportunity for microbial life on the more numerous colder bodies in the solar system. However, any ammonia solvent in this type of environment should not be expected to be pure. Water in the form of ice or icebergs would dissolve in liquid ammonia; thus, the occurrence of water groups such as OH<sup>–</sup> and O<sup>2–</sup> should be expected within an ammonia solvent, if it indeed is capable of supporting life. Any ammonia–water mixture would be very basic in nature (e.g., a 1.7% ammonia solution has a pH of 11.6; Budavari et al. (1996), though, and it is not clear how well life could adapt to this high basicity. On Earth, life is much more capable of withstanding and adapting to extreme acidities than basicities. However, there are not many natural environments on Earth with high pH values; thus, there may have been no need for the evolution of Terran organisms to develop this type of adaptation. Therefore, high pH may not be a fundamental obstacle for life.

#### Organic solvents

Non-polar hydrocarbons such as methane and ethane are abundant in the universe and are probably the most common solvent on one moon of our solar system (Titan). A hydrocarbon solvent would be beneficial to organic synthesis reactions without destroying hydrolytically unstable organic species including nucleobases (as water does). Thus, a hydrocarbon solvent may actually improve chances for the origin of life. Decades of experience with organic synthesis reactions has shown that the presence of water

greatly diminishes the chance of constructing nucleic acids and most other organic macromolecules. Broad empirical experience has demonstrated that organic reactivity in hydrocarbon solvents is no less versatile than in water, and many Terran enzymes are believed to catalyze reactions by having an active site that is not water-like (Benner et al. 2004). A hydrocarbon solvent would also provide protection against UV radiation, as hydrocarbon smog emanating from the liquid solvent would absorb some of the UV radiation and thus, offer it a significant degree of protection (Schulze-Makuch and Irwin 2004). At least one organism is known to be able to thrive in a hydrocarbon environment (Marcano et al. 2002). Organisms in a non-polar solvent, however, would have to have a biochemistry quite different from that of Terran organisms. For example, membranes of Terran organisms, which are submerged in the polar solvent, water, are amphiphilic with their polar (hydrophilic) heads immersed in the solvent and their non-polar (hydrophobic) tails oriented toward each other, away from the solvent. They interact with the polar solvent to take up nutrients, respond to intracellular signals, and discard wastes. If a non-polar solvent could support life, the chemical orientation might be reversed analogous to reverse micelles, or may consist of multi-layer lipids. Cellular membranes in an organic solvent might incorporate silanes as building blocks. Silanes are liquid while polysilanes are solid under Titan surface conditions (Table 2).

There are also hydrocarbon compounds that are polar, for example, methyl alcohol ( $\text{CH}_3\text{OH}$ ) in which an H is replaced with an OH group. This compound is an excellent polar solvent, a better temperature moderator than water, and has a larger liquidity range than water (Table 1). It may also be a relatively common compound in the universe as it has been found in the interstellar medium and in comets (Goldsmith and Owen 2001). Formamide is another polar organic compound that has been suggested. Benner et al. (2004) pointed out that many chemical species that are thermodynamically unstable in water with respect to hydrolysis are spontaneously synthesized in formamide. This includes ATP from ADP and inorganic phosphate, peptides from amino acids, and even oligoribonucleotides (Schoffstall and Liang 1985; Schoffstall et al. 1982). Hydrocarbons with polar groups can also segregate into immiscible phases, such as those of acetonitrile and hexane. Compartmentalization could, thus, be achieved in liquid/liquid phase separation in bulk hydrocarbon (Benner et al. 2004). A discussion of methane/ethane as a possible life-supporting solvent is provided below in the case of Titan's surface conditions.

**Table 2** Melting and boiling points of some silanes (modified from Greenwood and Earnshaw 1984)

Physical property	$\text{SiH}_4$	$\text{Si}_2\text{H}_6$	$\text{Si}_3\text{H}_8$	$n\text{-Si}_4\text{H}_{10}$
Melting point (°C/K)	-185/88	-133/140	-117/156	-90/183
Boiling point (°C/K)	-112/161	-14.3/259	53.1/326	108/381

## Other solvents

There are alternatives to water other than ammonia and organic solvents. Any liquid or supercritical fluid can be considered as a potential solvent, in principle. For example, even Terran enzymes can operate in a variety of not only non-aqueous solvents but completely apolar solvents such as benzene, diphenyl ether, or straight-chain hydrocarbons from heptane through hexadecane (Carrea et al. 1995; Klivanov 2001). Benner (2002) even suggested sulfuric acid ( $\text{H}_2\text{SO}_4$ ) as a possible solvent for Venus. On the surface of Venus, carbon dioxide is a supercritical fluid, which is an interesting medium for chemical and biochemical processes (e.g., Ikushima 1997). Bains (2004) suggested liquid nitrogen as an abundant and suitable solvent for very cold planetary bodies such as Triton (e.g., Soderblom et al. 1990) and hypothesized that silanols could dissolve in liquid nitrogen at concentrations sufficient to be precursors of life. Liquid nitrogen, though, has a very limited temperature range of liquidity. Most of the potential solvent candidates are liquids at lower temperatures than water (Table 1). Hydrazine,  $\text{N}_2\text{H}_4$ , which is liquid from 2 to  $114^\circ\text{C}$  at 1 bar pressure, is one of the few exceptions. There is, however, overlap with the thermal range of liquidity for water in some cases (e.g., HCN, HF). Chemical reactions occurring within these solvents would proceed at a much slower pace than on Earth, typically by a factor of 2 for every  $10^\circ\text{C}$  decrease in temperature (Jakosky 1998). However, the key to developing a suitable chemistry at a given temperature lies in selecting chemical reactions suited to that temperature (Schulze-Makuch and Irwin 2004). The liquidity range of a variety of other solvents is shown in Fig. 1, and their physical properties are illustrated in Table 1.

## Energy sources for life

Life has evolved on Earth with light, organic molecules, and oxidizable inorganic chemicals as abundant sources of energy. Thus, living systems have adapted to the use of these forms of energy, though a broader range of potential energy sources is actually available. Organisms can sense thermal energy (Viswanath et al. 2003), pressure, stretch, movement of water and air, gravity, electric and magnetic fields (Blakemore and Frankel 1981; Frankel et al. 1979; Schmid-Nielsen 1990), and a wide range of chemical substances. All these stimuli ultimately generate responses through membrane transductions, either by altering the gating of ions that change transmembrane potentials or by initiating metabolic changes through secondary messenger systems (Schulze-Makuch and Irwin 2004). Thus, they point out the capacity for energy in a variety of forms to affect biological processes (Schulze-Makuch and Irwin 2002a).

Energetically favorable redox reactions are the basis for life on Earth. The best known and most common types of metabolism are based on hydrogen oxidation and methanogenesis, sulfur reduction and oxidation, iron and

manganese reduction, denitrification, and aerobic respiration. However, many other energy-yielding redox reactions are known that involve the reduction or oxidation of relatively rare elements such as arsenic, selenium, copper, lead, and uranium. Thus, there does not appear to be a basic limitation on which elements or redox reactions can be used. Rather, the reactions that occur will likely be dictated by the abundance, availability, and suitability of a specific element in a certain type of environment. The diversity of arsenic and selenium available to respiring bacteria thriving in playas (alkaline salt lakes) and mining tailings is a good example (Stolz and Oremland 1999).

Probably, one of the most important factors in the success of life on Earth is the use of a narrow band of light as an energy source. Without the use of light, life on Earth would be much more limited in abundance and diversity. The evolution of photosynthesis allowed life to tap into a practically unlimited source of energy. Sometime early in the history of the Earth, photosynthesis developed as a favorable energy-capture mechanism. The frequency varies somewhat for different organisms depending on which wavelength is used for photosynthesis. For example, bacterial chlorophyll uses a wavelength of 800 to 1,000 nm, while carotenoids in plants absorb wavelengths ranging from 400 to 550 nm. The average energy gain across the visible spectrum is about 190 kJ/mol. It is interesting to note that this value is very similar to the free energy provided by hydrogen-oxidizing prokaryotes thought to represent early chemoautotrophic organisms (Schulze-Makuch and Irwin 2004). Thus, from a purely energetic viewpoint, light energy and chemical energy are equally competitive.

#### Electromagnetic wavelengths not known to be used by Terran organisms

The wavelength of light used by phototrophic organisms lies in the visible and near-infrared spectrum. The fact that these specific frequencies are used may merely reflect adaptation by Terran organisms to the most prevalent wavelengths of electromagnetic radiation emitted from the Sun. These wavelengths are also transmitted through our atmosphere well, making them readily available to life on the surface of our planet. However, in theory, other wavelengths could be harvested. Scavenging photons that are richer in energy would be an advantage in principle, but organisms would have to develop a control mechanism to avoid being overwhelmed by the influx of energy. The problem with more energy-rich radiation such as that of ultraviolet (UV) light is that it is detrimental to many life-supporting molecules such as proteins and DNA (Rettberg and Rothschild 2002). Thus, any organism using UV or more energy-rich radiation would need some kind of protection against damaging wavelengths to harvest this type of energy, as may be the case for possible life in the Venusian atmosphere, as described below. Otherwise, the organism would have to be based on a very different type of biochemistry. Alternatively, organisms may be able to

make use of less energy-rich radiation, which is plentiful in the universe, as any body or substance above 0 K will radiate infrared light. A prime example is provided by hydrothermal vents on the ocean floor of Earth, where the only source of light is geothermal radiation that includes wavelengths that can be absorbed by photosynthetic pigments of organisms. The recent capture of such an organism, a green sulfur bacterium, at a deep-sea hydrothermal vent, but not from surrounding waters, indicates that geothermal light and associated reduced sulfur compounds are sufficient to at least enhance the survival of green sulfur bacteria in the otherwise dark, oxygenated depths and expands the range of possible environments that could harbor life forms which use light energy to drive endergonic biochemical reactions (Beatty et al. 2005). However, more photons would need to be harvested to yield enough energy to be biologically useful. For example, one photon of infrared radiation would, in most instances, not supply enough energy to make the terminal ATP bond. Also, Gusev and Schulze-Makuch (2005) recently provided experimental evidence that microbes may be capable of converting radio waves into chemical energy to supplement their energy needs. They hypothesized that protons in liquid water could be excited at their natural resonance frequencies by the Sun's and Earth's natural magnetic field through Langmuir oscillations, generating enough kinetic energy to charge the transmembrane potential of a cell.

#### Thermal energy gradients

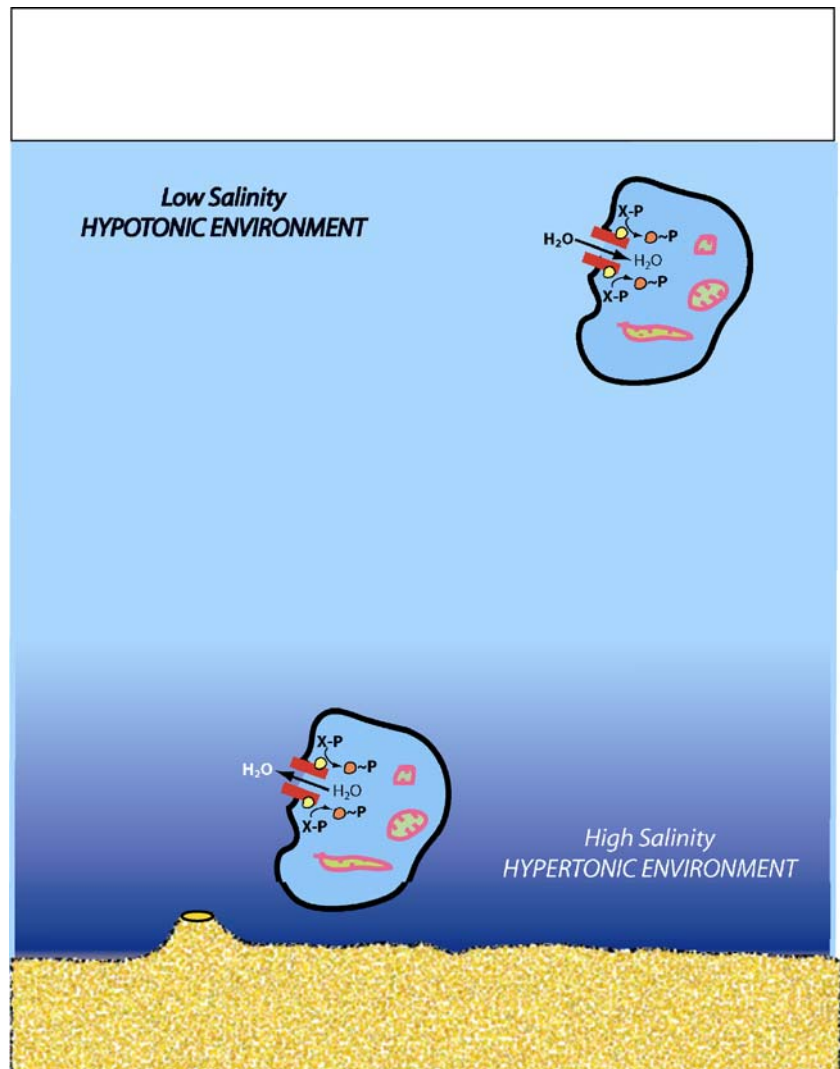
Thermal gradients could, in principle, also be used by organisms to obtain energy. Muller (1985) envisioned a theoretical mechanism that would allow biomembranes to convert heat into electrical energy during temperature cycling. This process of thermosynthesis could possibly be used by organisms living in convecting volcanic hot springs. Thermosynthesis, just like a steam engine, would make use of a phase transition. Membranes can undergo the thermotropic phase transition, which increases the mobility of the molecules within the membrane (Muller and Schulze-Makuch 2005). Due to a change in dipole potential, such a transition would very plausibly result in a change in potential across the membrane (Muller 1993). Although this potential change has not been measured directly, similar potential changes that undergo the thermotropic phase transition have been measured across monolayers of lipids at the water/air interface. The changes can easily reach 100 mV, high enough to drive ATP synthesis. In his model, electrogenic ATPases would convert the electrical energy gained by thermosynthesis into ATP if their activity and stoichiometry were properly regulated (Muller 1993). If correct, thermosynthesis could be a plausible basic pathway of metabolism for early organisms on Earth, possibly a progenitor of bacterial photosynthesis (Muller 1995, 2003), and an option for possible life on other worlds.

## Osmotic gradients

Osmotic gradients can also be an enormously powerful source of energy. Some halophilic strains of cyanobacteria are known to tolerate salt concentrations of up to 2.7 M NaCl (Hagemann et al. 1999). Marine teleosts (bony fish such as shark) retain a strong osmotic differential of roughly 0.7 Osm between their intercellular fluids and their surrounding environment (Wilmer et al. 2000), where 1 Osm is one mole of osmotically active particles. Schulze-Makuch and Irwin (2004) used this conservative figure, assuming water molecules moving through a membrane channel that couples the movement to the formation of a high-energy covalent bond, and calculated the potential energy yield to be sufficient for phosphorylating one ATP molecule from ADP for about every 45 water molecules entering the cell by osmosis. This energy gain was based on those of fish that have adapted from their freshwater origin to their marine environment. Halophilic microbes that tolerate much more extreme osmotic gradients might easily be able to more than quadruple this energy yield. While the direct coupling of water movement to phosphorylation re-

actions is unknown for living systems on Earth, evolution under circumstances where osmotic gradients provide free energy could have favored the origin of membranes in which water movement causes tertiary structural changes in a channel-associated protein that catalyzes formation of high energy bonds, such as ligand-induced conformational changes in membrane receptors lead to a series of steps culminating in the synthesis of high-energy cyclic AMP (Schulze-Makuch and Irwin 2002a). A drawback to the use of osmotic gradients for generating free energy is the fact that the influx of many water molecules would significantly increase the cell volume. This could be mitigated, however, by a compensatory loss of solutes, such as efflux of  $\text{Na}^+$  (and  $\text{Cl}^-$  for electrical balance) powered by the rise in intracellular pressure or simply by salt precipitation. To regenerate the osmotic gradient that would be depleted, an osmotrophic organism could move between a hypertonic ocean floor and hypotonic water some distance above the ocean floor. The zone close to the ocean bottom would be expected to be high in total dissolved solids due to a higher density. Strong osmotic gradients would be present in both solutions but in opposite

**Fig. 2** A hypothetical osmotrophic organism that harvests energy from salinity gradients. Movement of water in either direction would be coupled to a reaction that forms a high-energy covalent bond ( $\sim\text{P}$ ) by a membrane molecular complex energized by entrance or exit of water (modified from Schulze-Makuch and Irwin 2004)



directions if not much water circulation is occurring. For example, a membrane water channel could be coupled to a reaction that forms a high-energy bond inside the cell as the water moves inward from hypotonic surroundings, while a similar channel oriented in the opposite direction could harvest energy when water leaves the cell in hypertonic surroundings. The hypothetical organism could thus, move between two layers of different salinity, using both to harvest energy (Fig. 2). Thus, the harvest of osmotic gradients on other planetary bodies is a reasonable possibility, especially when strong chemical concentration gradients are present in the environment and other energy-yielding strategies may not be feasible.

### Electromagnetic fields

Charge separation and extractable free energy can also be generated from magnetic fields, in theory. Magnetic fields can yield energy based on the Lorentz force, the movement of a charge within a magnetic field, or by induction from a periodically changing magnetic field. A plausible mechanism for harvesting energy from magnetic fields is provided by  $H^+$  ions that are driven across a one-way channel against their concentration gradient into an internal organelle, where they accumulate to a higher concentration gradient than on the outside of the organelle. Then, the  $H^+$  ions can diffuse back out of the organelle through a different channel coupled to a reaction that produces a high-energy covalent bond (similar to how mitochondria generate high-energy organic phosphates; Schulze-Makuch and Irwin 2004). A more elaborate scheme would be the use of the Lorentz force to separate one electron and one proton from the center of the cell in opposite directions to the respective ends of the hair cell. When the magnetic field lines are oriented parallel to the hair cell, the energy could be released and the proton and electron would move toward each other producing  $H_2$  (Fig. 3).

The amount of energy that can be extracted via the Lorentz force depends on the strength of the magnetic field of the particular planetary body. The strength of Earth's magnetic field or even Jupiter's much stronger field do not

provide enough energy to make magnetic energy compatible with chemical or light energy on Earth. However, while terrestrial magnetic fields are not powerful enough to drive biochemistry, other magnetic fields that exist in nature might be. For example, the neutron star SGR 1806-20, a magnetar, has been reported to have a magnetic field strength of about  $10^{15}$  G ( $10^9$  T) (Ibrahim et al. 2003). Energies from these huge fluctuating magnetic fields could become competitive for organisms on planets orbiting neutron stars, depending on the magnetic field strength of the particular star, distance of the planetary body from the neutron star, environmental conditions on the planet or moon, and the availability of suitable niches below the planetary surface to protect from radiation intensity, among other factors.

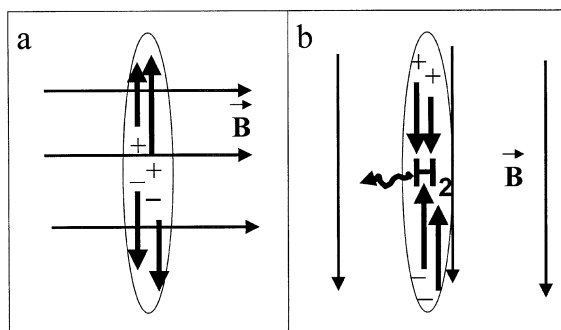
Other energy sources are also available, in principal, for supporting biological activity. For a detailed discussion of these other possibilities the reader is referred to (Schulze-Makuch 2002a and Irwin 2004).

### A selection of plausible habitats for life without precedent on earth

After elaborating on biochemical principles, let us discuss some habitats where life would not be expected to be present by the standards of conventional Terran biology. While the possibility for microbial life in the Martian subsurface and the European subsurface ocean are well accepted, the sites introduced here are not. However, based on considerations of planetary habitability and our definition of life, we feel that these sites have sufficient potential to merit a closer look. We discuss the atmosphere of Venus, a site possibly relevant to many other planetary atmospheres, and the surface of Titan, indisputably one of the most exotic places in our solar system.

#### Life in the atmosphere of Venus?

Assuming the presence of liquid water on the surface of Venus during its early history (e.g., Abe and Matsui 1988; Donahue et al. 1982) and given its dynamic endogenic-driven activity, it seems reasonable that life could have originated on Venus. Another possibility is that organisms could have been delivered by meteorites from early Earth or Mars to a suitable habitat on early Venus. Recent studies suggest that a significant amount of bacteria can survive space travel for millions of years, including lift off from the place of origin and re-entry into a planetary atmosphere (Davies 1996; Horneck and Rettberg 2002). Life could then have retreated into ecological niches progressively through natural selection during the period of greenhouse warming (Schulze-Makuch and Irwin 2002b). Whether the adaptation of full life cycles to an exclusively atmospheric habitat can occur is unclear. However, if the run-away moist greenhouse effect occurred gradually rather than catastrophically, adaptation to life in a thick and nutrient-rich atmosphere appears feasible. The possibility of life in



**Fig. 3** Life scheme for magnetotrophic organism. **a** Lorentz force separates protons and electrons. **b** Magnetic field lines are oriented parallel to long axis of microbe and protons and electrons form molecular hydrogen (from Schulze-Makuch 2004)

the Venusian atmosphere has been discussed by various authors (Cockell 1999; Grinspoon 1997; Morowitz and Sagan 1967; Sagan 1961; Schulze-Makuch and Irwin 2002b; Schulze-Makuch et al. 2004). Their arguments can be summarized as follows: (1) the clouds of Venus are much larger, more continuous, and stable than the clouds on Earth; (2) the atmosphere is in chemical disequilibrium, with  $\text{H}_2$  and  $\text{O}_2$ , and  $\text{H}_2\text{S}$  and  $\text{SO}_2$  coexisting; (3) the lower cloud layer contains non-spherical particles comparable in size to microbes on Earth; (4) conditions in the clouds at 50 km in altitude are relatively benign, with temperatures of 300–350 K, pressure of 1 bar, and a pH of about 0; (5) the super-rotation of the atmosphere enhances the potential for photosynthetic reactions; (6) COS is present in the atmosphere, which on Earth is a strong indicator of biological activity; (7) CO is less abundant than expected under Venusian atmospheric conditions and could be oxidized as a reactant in plausible metabolic pathways (see Eqs. 2 and 3 below); (8) the biologically critical elements of carbon, phosphorus, and nitrogen are present; and (9) while water is scarce on Venus, water vapor concentrations reach several hundreds of parts per million in the lower cloud layer.

The major problems confronting possible microbial life in the Venusian atmosphere would be the lack of water, the low pH, and the large amounts of UV radiation that the Venusian atmosphere receives. The lack of water could be overcome by microbial organisms if they have developed a mechanism with which they assimilate water vapor from hydrated sulfur compounds or from the atmosphere, similar to the assimilation of carbon from  $\text{CO}_2$  by microbes in the atmosphere of Earth. Although a pH of about 0 in the lower cloud layer may seem extreme, Schleper et al. (1996) isolated microorganisms on Earth that thrive at this pH

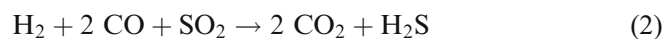


**Fig. 4** Ultraviolet image of Venus' clouds as seen by the Pioneer Venus Orbiter (5 February 1979). The *black streaks* are thought to result from an unknown UV absorber material, which could be related to microbiological processes in the Venusian atmosphere (source NSSDC)

level. Nevertheless, a low pH constitutes a problem due to the instability of the nucleotides adenine and cytidine at low pH. Earth organisms that live at low pHs have adapted to this problem by pumping protons to maintain an intracellular pH well above the  $\text{pK}_a$  (negative logarithm of the equilibrium coefficient of the neutral and charged forms of a compound, also referred to as acid ionization constant) of the protonated nucleobases. A more effective adaptation of microbial life on Venus could be to replace cytidine with a base such as 5-nitrocytidine, which has a considerably lower  $\text{pK}_a$  than cytidine (Benner et al. 2004). With regard to the high amount of UV radiation in the Venusian atmosphere, cycloocta sulfur ( $\text{S}_8$ ) could be used by microbes for protection (Schulze-Makuch et al. 2004).  $\text{S}_8$  has the capability of shielding organic macromolecules such as DNA and protein at wavelengths most susceptible to UV damage.  $\text{S}_8$  is also thermodynamically stable and does not react with sulfuric acid. In fact, an unknown UV absorber has been detected in the Venusian atmosphere (Fig. 4), and Hapke and Nelson (1975) concluded that  $\text{S}_8$ , along with some polymorphic sulfur, provides the best match. If  $\text{S}_8$  is used as a kind of “sunscreens,” it would explain why only minute amounts of organic compounds were detected by spectral methods (e.g., Plummer 1969). A somewhat analogous process is observed on Earth, where some purple sulfur bacteria, green sulfur bacteria, and some cyanobacterial species deposit elemental sulfur granules outside of the cell (e.g., Pierson et al. 1987; Tortora et al. 2001). Also, recent investigations of thermophilic microbes in Yellowstone Park, Wyoming revealed microbial filaments fully covered by a mineral phase that contains significant amounts of elemental sulfur (McDermott, personal communication). Schulze-Makuch and Irwin (2002b, 2004) suggested the possibility of phototrophic life at Venus based on Photosystem I. Anoxygenic photosynthesis commonly occurs in anaerobic near-surface environments on modern Earth. Many organisms using this photosystem thrive in warm seas, soils, and hot springs (e.g., Bryantseva et al. 2000, Vethanayagam 1991) and may have been ideal inhabitants for a warm proto-ocean on early Venus.



Alternatively, Schulze-Makuch and Irwin (2002b) also suggested possible metabolic pathways for chemotrophic organisms that would be reasonable under environmental conditions prevailing in the current Venusian atmosphere:



The Gibbs free energy yielded would be larger than 240 kJ/mol for either pathway (Eqs. 2 and 3). Thus, the

presence of microbial life in the current Venusian atmosphere should be considered as a distinct possibility (Schulze-Makuch et al. 2005).

Life in the atmosphere may be more common than we might expect. There are some indications that microbial growth and reproduction occurs in Earth's atmosphere. Dimmick et al. (1979) reported the division of bacteria on airborne particles. More recently, Sattler et al. (2001) analyzed condensing clouds at the Sonnblick Observatory in Austria at an altitude of 3,106 m and suggested growth and reproduction of microbes in super-cooled cloud droplets. It has been recognized for some time that bacteria exist in cloud aerosols on Earth (e.g., Gislén 1948) and that rain and fog water rich in nutrients may provide a good substratum for microorganisms (Fuzzi 2002; Herlihy et al. 1987). While the dispersal of microorganisms by wind is well accepted (Schulze-Makuch 2003) and may even occur within the stratosphere (Imshenetsky et al. 1978), the claim that microbes independently grow and reproduce in Earth's atmosphere is controversial. However, in general, the atmosphere of Earth is a poor analogy for atmospheric habitats where life would be more likely, namely, that of planetary bodies or satellites where gases are denser and liquids are found in larger aggregates with longer survival times (Schulze-Makuch and Irwin 2004).

#### Life on the surface of Titan?

Environmental conditions are generally thought to be conducive for life if it can be shown that (1) polymeric chemistry, (2) an energy source, and (3) a liquid solvent are present in appreciable quantities (Irwin and Schulze-Makuch 2001). This is the case for Titan. The presence of large methane and ethane reservoirs with traces of dissolved N<sub>2</sub> on Titan's surface has long been suspected (Lunine 1994) and the outgassing of methane from the surface at the landing site of the Huygens probe is consistent with this assumption. Recent Cassini radar images and visible images from the Huygens lander suggest the presence of a young surface and ongoing cryovolcanism (Elachi et al. 2005; Porco and The Cassini Imaging Team 2005), which points toward near-surface liquid reservoirs and strongly implies at least occasional surface flows of liquid methane or possibly water–ammonia mixtures. In addition, abundant energy sources are present in the form of UV radiation and high-energy molecules produced from photochemistry.

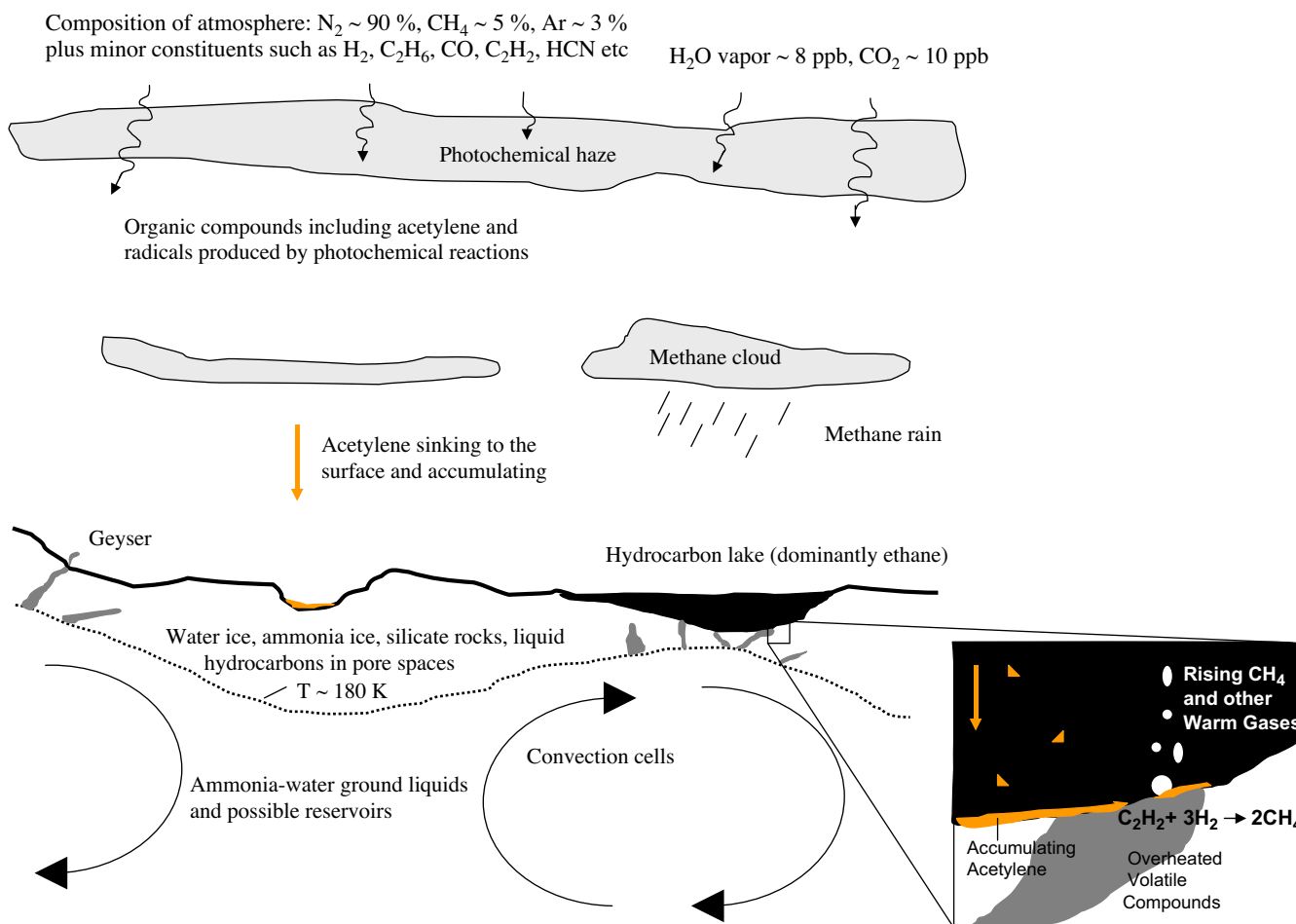
The presence of liquid methane presumably improves the chances for the origin of life because the assemblage of organic macromolecules that could give rise to life appears to be more straightforward in a hydrocarbon environment. The biochemistry of an organism in a hydrophobic solvent would be quite different though, as discussed above (see under “Organic solvents”). Also, given Titan's extremely low availability of unbound oxygen, O molecules might be replaced with N analogues as suggested by Molton (1974) and more recently by Raulin and Owen (2002). Furthermore,

the miniaturization of cellular life in water on Earth may be a misleading model for life in a non-aqueous environment (Schulze-Makuch and Irwin 2004). In an extremely cold, hydrophobic (but liquid) environment, surface to volume ratio considerations may be less constraining than at higher temperatures in polar solvents because of the lower viscosity of the solvent (Table 1) and the slower diffusion rates permitted by the greatly reduced rate of metabolism. Thus, life on Titan could involve huge (by Earth standards) and very slowly metabolizing cells.

A methane cycle may exist on Titan with some similarities to the hydrological cycle on Earth (Baker et al. 2005; Schulze-Makuch and Grinspoon 2005). Methane clouds have recently been detected (Porco and The Cassini Imaging Team 2005; Roe et al. 2002), and methane rain is consistent with modeling results (Chanover et al. 2003; Tokano et al. 2001). Fortes (2000) pointed out that given the extremely cold surface, some of the simplest prebiotic reactions on Titan would have half-lives on the order of 10<sup>7</sup> years. Perhaps, however, reactions could be accelerated by catalysis or localized warming. Regions of geothermal activity have been projected to exist on Titan (Lorenz 2002), and the early Cassini results suggesting the presence of a young surface with widespread cryovolcanism (Elachi et al. 2005; Porco and The Cassini Imaging Team 2005) are suggestive of periodic heat-driven processes.

As an alternative to metabolism in organic solvents, the possibility of a water-based metabolism more similar to that of living systems on Earth cannot be discounted. Water droplets within a hydrocarbon environment could provide convenient cellular compartments for molecular evolution (Tawfik and Griffiths 1998). An emulsion of water droplets in hydrocarbons is obtainable simply by shaking (Benner et al. 2004). While Titan's current surface temperature is very cold (~94 K), early in its history Titan would have been likely exposed to a large amount of greenhouse warming, with accretional and radiogenic heat having kept it much warmer than today. Water–ammonia mixtures will remain liquid to temperatures as low as 176 K (e.g., Croft et al. 1988), allowing amino acids and other macromolecules from space and those produced by atmospheric photochemical processes to interact with potential solvents. Ammonia–water volcanism and meteorite impacts may have created episodes of aqueous chemistry in lakes on Titan's surface, perhaps lasting thousands of years before freezing over (Artemieva and Lunine 2003; Thompson and Sagan 1992). An especially promising environment for life would thus be a hot spring or geothermal area at the bottom of a hydrocarbon reservoir or an area where volatile overheated compounds intersect with such a reservoir (Fig. 5). This environment would not only provide a versatile suite of raw material for organic synthesis and some amount of molten water and ammonia for organic reactions but also higher temperatures for reactions to occur more rapidly (Schulze-Makuch and Grinspoon 2005).

Given the environmental conditions on Titan, a reasonable energy-yielding reaction for a metabolizing microbe



**Fig. 5** Schematic of environmental conditions at Titan (modified from Schulze-Makuch and Grinspoon 2005). Acetylene and radicals are produced by photochemical reactions in the atmosphere. Because of its high specific gravity, acetylene will sink to Titan's surface and to the bottom of a hydrocarbon reservoir, where it can be used by putative organisms for metabolic reactions (*inset*)

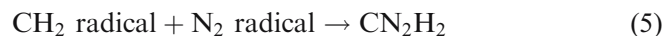
would be the catalytic hydrogenation of photochemically produced acetylene (Abbas and Schulze-Makuch 2002; Schulze-Makuch and Grinspoon 2005):



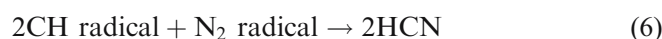
The energy yield of this reaction is  $107.7$  kJ/mol ( $\Delta G = -107.7$  kJ mol $^{-1}$  or  $-25.7$  kcal mol $^{-1}$ ) under standard conditions and about  $100$  kJ/mol under Titan's surface conditions. Both acetylene and hydrogen are present in Titan's atmosphere at significant concentrations. As acetylene is produced high in the stratosphere from solar UV radiation and then, for the most part, condenses and falls to the surface, it provides a potential means of transferring high-altitude solar UV energy to surface chemical reactions (Lorenz et al. 2000). Spectroscopic evidence suggests that the product of this reaction, methane, is found to be isotopically lighter than would be expected from theories of Titan's formation (Lunine et al. 1999) and, thus, may hint toward microbial fractionation.

Other metabolic pathways are possible on Titan as well. Smith and McKay (2005) suggested, in addition to acetylene, the endogenic reactions of ethane and organic solids with atmospheric hydrogen as possible metabolic pathways, and concluded that methanogenic life in liquid methane on Titan may be widespread.

Another intriguing possibility is the use of free radical reactions as a basis for metabolism. Raulin (1998) suggested that Titan's stratosphere is an active site of complex carbon and nitrogen radical chemistry. Thus, energy-yielding reactions could be based on chemistry involving radicals. For, example, a chemoautotrophic organism may use the following reactions:



or



Reactions 5 and 6 produce a high yield of energy and could take place in the atmosphere or at the surface of Titan. All reactants have been detected in Titan's environment based on data from Voyager 1 (Kunde et al. 1981; Smith et al. 1982). On Earth, energy-rich reactions involving radicals are very difficult to control and would cause internal damage to any organism. On Titan, however, at surface temperatures of less than 100 K, these reactions may proceed at a reasonable pace, and may constitute a feasible energy-yielding reaction for a metabolic pathway. An interesting consequence of the radical reactions 5 and 6 is the production of the biologically important compounds cyanamide and hydrocyanic acid, respectively. Various pathways have been proposed for assembling amino acids and proteins using cyanamide and HCN as basic compounds (e.g., Abbas and Schulze-Makuch 2002).

As discussed before, the recent Cassini–Huygens results indicate that Titan has an extremely young and active surface, with preliminary crater counts (from incomplete surface imaging) that suggest a surface age of less than 300 million years (e.g., Elachi et al. 2005; Porco and The Cassini Imaging Team 2005) and many features that suggest the likelihood of cryovolcanic activity. Titan has a low bulk density of  $1.88 \text{ g cm}^{-3}$ , which would imply that silicate substrate is quite rare at the surface with less heat produced by radioactivity. Thus, the observed smoothness and activity is somewhat puzzling. An intriguing, though highly speculative, possibility has recently been advanced by Schulze-Makuch and Grinspoon (2005). They suggested that biological heating could contribute to the surface activity and smoothness. Energy-rich organics, such as acetylene, are created in the upper atmosphere, by the interaction with solar radiation and the electromagnetic field of Saturn, and fall to the surface. Through geological overturn and meteorological/fluvial processes, these compounds are transported to the subsurface where they can reach liquid reservoirs and serve as the basis for metabolism, using the reactions detailed above. Some of the energy from these highly exothermic metabolic reactions would be released to the surroundings and, in some circumstances, contribute to melting of water–ammonia ice (Fig. 5). Glacial melting in Earth near polar regions from biothermal energy released by algal metabolism has been reported previously (Gerdel and Drouet 1960), as has been the influence of marine microorganisms on the melting of Arctic pack ice (Leck et al. 2004). Microbial colonization occurs in cryoconite holes on glaciers (Wharton et al. 1985) and in basal glacial melt waters, some of which are known to release methane as a metabolic end product (Campen et al. 2003; Souchez et al. 1995). In an environment near the freezing point where availability of liquid microenvironments is a limiting factor on habitability, there may be selective pressure leading to a larger portion of the metabolic heat going into the environment. There may be places on Titan that are energy rich but liquid poor, with plenty of acetylene to metabolize, though it may be locked up in ice. In such a case, evolution would favor organisms that could use this energy to melt their own little watering

holes. Schulze-Makuch and Grinspoon (2005) provide calculations that this strategy could work, in principle.

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## Discussion

The major purpose of this review has been to argue the possibility that alien life on other worlds may be very different from the example provided by the only life we know with certainty, as it occurs on our home planet. Without resorting to physical laws or chemical principles that differ from those presumed to apply universally, we have argued that organisms, unlike any known on Earth, could well exist within our own solar system. As examples, we have pointed to the possibility of extremely acidophilic microbes residing in water droplets in the upper atmosphere of Venus and of exotic organisms using either organic solvent-based metabolism or water-based metabolism encased within a hydrocarbon environment on Titan. This does not, of course, preclude the strong possibility that carbon-based life, perhaps derived from a common origin somewhere among the rocky planets of the inner solar system or originating and evolving in somewhat parallel lines anywhere that liquid water habitats can be found, would not be recognizably similar to life as known on Earth. We have simply sought to emphasize that life could exist elsewhere, either as we know it or in a form totally unknown to us.

Even within the one sample of life that we have, unanswered questions confound our ability to predict the course that life might take elsewhere. We do not know, for example, whether macroorganisms require a highly efficient form of energy metabolism, as provided by aerobic respiration, in order to grow beyond the microbial level. Why did macroscopic life arrive so late in the history of the Earth? Is an oxygenated atmosphere really needed or was that only a special case for Earth? How did the first cells originate? How did the undirected chaos of protometabolic systems come under the control of complex molecules synthesized with high fidelity by template-guided directions? These are all mysteries awaiting clarification and, until they are answered, a complete understanding of even the single case of life with which we are familiar will remain elusive. If and when we discover life on other worlds, whether it be like the life we know or totally exotic in substance as well as alien in origin, astrobiology will have become an observational rather than a speculative science. As such, it will rank among the most exciting discoveries of all time.

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