Sustainability **2010**, *2*, 1602-1623; doi:10.3390/su2061602

OPEN ACCESS

sustainability

ISSN 2071-1050

www.mdpi.com/journal/sustainability

Review

Resistance of Microorganisms to Extreme Environmental Conditions and Its Contribution to Astrobiology

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Received: 26 April 2010; in revised form: 6 May 2010 / Accepted: 24 May 2010 / Published: 4 June 2010

Abstract: In the last decades, substantial changes have occurred regarding what scientists consider the limits of habitable environmental conditions. For every extreme environmental condition investigated, a variety of microorganisms have shown that not only can they tolerate these conditions, but that they also often require these extreme conditions for survival. Microbes can return to life even after hundreds of millions of years. Furthermore, a variety of studies demonstrate that microorganisms can survive under extreme conditions, such as ultracentrifugation, hypervelocity, shock pressure, high temperature variations, vacuums, and different ultraviolet and ionizing radiation intensities, which simulate the conditions that microbes could experience during the ejection from one planet, the journey through space, as well as the impact in another planet. With these discoveries, our knowledge about the biosphere has grown and the putative boundaries of life have expanded. The present work examines the recent discoveries and the principal advances concerning the resistance of microorganisms to extreme environmental conditions, and analyzes its contributions to the development of the main themes of astrobiology: the origins of life, the search for extraterrestrial life, and the dispersion of life in the Universe.

Keywords: astrobiology; extremophiles; origins of life; Mars; Europa; panspermia

1. Introduction

Living organisms tend to be sensitive to drastic changes in their environments. Extreme conditions of temperature, pressure, drought, salinity, and pH disrupt the crucial interactions that keep biomolecules folded and functional, thereby quickly destroying the cellular integrity [1]. Therefore, scientists have tended to assume that there are strict boundaries to the biosphere imposed by terrestrial life's requirement for a rather narrow and specific range of environmental conditions.

Discoveries in the past few decades, however, have shown that life is not always as sensitive as we might have imagined, and that the limits of life on Earth are far from being well defined [2]. Historical notions of what is or is not a hostile environment have turned out to be erroneous.

For every extreme environmental condition investigated, a variety of microorganisms have shown that they not only can tolerate these conditions, but that they also often require those conditions for survival [3]. Extreme environmental conditions may be considered natural or simulated forces, which make difficult the survival and development of most living systems. Such conditions include physical extremes (e.g., temperature, radiation, and pressure) and geochemical extremes (e.g., desiccation, salinity, pH, and redox potential) [4]. In addition to the familiar metabolic pathway of photosynthesis, microbes possess metabolisms based upon methane, sulfur, and even iron [5]. If this wide and versatile metabolic diversity is coupled to the extraordinary physiological capacities of many microorganisms to colonize extreme environments, it becomes evident that there is an unlimited amount of habitats on Earth that are suitable for microbial life.

Microorganisms are found to thrive at 6.7 km depth inside the Earth's crust, and more than 10 km deep inside the ocean at pressures of up to 110 Mpa; from extreme acid (pH 0) to extreme basic conditions (pH 12.8); and from hydrothermal vents at 122 °C to frozen sea water at −20 °C. Recent developments in space technologies permit the study of adaptations and survival of terrestrial organisms to a new category of extreme conditions in space, including simulations of meteoritic impact or space conditions.

With these discoveries, our knowledge about the biosphere has grown and the putative boundaries of life have expanded [6]. In the last few decades, we have witnessed substantial changes in what scientists consider the limits of habitable environmental conditions.

Exploring the diversity of microorganisms and understanding their mechanisms of adaptation permit the development of hypotheses regarding the conditions required for the origin and early diversification of life on Earth [7]. The research also expands our notions of the potential habitable environments able to sustain life beyond Earth. Indeed, our increasing knowledge about the biology of microbes in extreme habitats has led numerous scientists to raise the possibility of finding life in various planetary bodies within the Solar System, including Mars, Venus, and the satellites of Jupiter (Io, Europa, Ganymede, and Callisto) and Saturn (Titan and Enceladus) [8,9]. Furthermore, a variety of studies in this field of research has given support to Panspermia—the hypothesis that life migrates naturally through space [10].

The present work examines the recent discoveries and the principal advances concerning the resistance of microorganisms to extreme environmental conditions and analyzes these studies' contributions to the development of the main themes of astrobiology: the origins of life, the search for extraterrestrial life, and the dispersion of life in the universe.

2. Microorganisms in Extreme Environmental Conditions

In this section we explore the diversity of extremophilic microbes, their mechanisms of adaptation, and examples of extreme environmental conditions where they are found.

2.1. Thermophiles and Hyperthermophiles

Thermophiles are microorganisms that thrive at relatively high temperatures, between 45 °C and 80 °C. Hyperthermophiles are particularly extreme thermophiles for which the optimal temperatures are above 80 °C [11]. Such microbes are found in various geothermally heated regions on Earth, such as volcanic soils permeated by hot vapors and deep-sea hydrothermal vents [12].

The best known and well-studied geothermal areas are in North America (Yellowstone National Park), Iceland, New Zealand, Japan, Italy, and the former Soviet Union [13]. These locations are usually rich in reduced chemicals from inside the Earth, and hence, many thermophiles are chemoautotrophic, reacting hydrogen, ferrous iron, or reduced sulfur compounds with electron acceptors like oxygen or nitrate [14]. As a consequence of extracting energy by oxidizing sulfur compounds, these reactions produce sulfuric acid, thus often making the geothermal waters very acidic. Consequently, many heat-loving microbes are also adapted to highly acid conditions. Indeed, hyperthermophilic extreme acidophiles, with pH optima for growth at or below 3.0 (like the archaea genera *Sulfolobus*, *Sufurococcus*, *Desulfurolobus*, and *Acidianus*), produce sulfuric acid from the oxidation of elemental sulfur or sulfidic ores in solfataras of Yellowstone National Park [15]. The most extreme examples of thermophilic, acid-loving extremophiles are two species of *Picrophilus*, which are able to grow at pH 0.7 and a temperature of 60 °C [16]. They were isolated from volcanically heated dry soils in Japan.

Deep sea hydrothermal vent communities are found near subsurface volcanoes and at the boundary between seawater and magma, usually kilometers beneath the ocean surface [17]. Since no light is available and the content of oxygen is very low, a large majority of thermophilic isolates from these deep sea locals are chemoautotrophic anaerobes. The present record of high-temperature growth is held by two Archaea isolated from these environments: Strain 121 and *Methanopyrus kandleri* can survive and reproduce at 121 °C and 122 °C, respectively [18,19].

The molecular basis for adaptations to extreme environments has been studied more intensively for high temperatures than for any other parameter. At high temperatures, biomolecules, such as enzymes, denature, losing their function and hence, stopping the metabolism. Also, the fluidity of membranes increases significantly, disrupting the cell.

To prevent denaturation and degradation, thermophiles present a variety of cellular adaptations. Their membrane lipids contain more saturated and straight chain fatty acids than do mesophiles (which grow typically between 15 °C and 40 °C) [20]. This allows thermophiles to grow at higher temperatures by providing the right degree of fluidity needed for membrane function. Thermophilic proteins appear to be smaller and in some cases more basic, which may also result in increased stability [21]. Another method used to improve the stability of proteins is through the action of chaperones, which help to refold denatured proteins [22]. Furthermore, monovalent and divalent salts enhance the stability of nucleic acids because these salts screen the negative charges of the phosphate groups, and KCl and $MgCl_2$ protect the DNA from depurination and hydrolysis [23]. Another way to stabilize DNA is through the employment of DNA-binding proteins and the compaction of the genome into chromatin [24].

2.2. Psychrophiles

Psychrophiles are microorganisms that grow at or below 0 °C and which have an optimum growth temperature of 15 °C and an upper limit of 20 °C [25]. They are found in a variety of cold environments, from the stratosphere to the deep-sea.

Most of the deep sea is at a constant temperature of 2 °C, while around the polar ice caps, liquid seawater may even be cooled to below 0 °C, as the typical salt content of sea water (3.4%) lowers the freezing point to −1.8 °C [26]. When the seawater freezes, the salt becomes increasingly concentrated in small pockets. Under these conditions, the freezing point of water may be depressed to −20 °C [27]. At such frozen temperatures, natural microbial metabolism has been measured. For example, the bacterium *Psychrobacter cryopegella* can grow at −10 °C, stay alive, and even keep metabolizing at −20 °C [28]. For this reason, many psychrophiles are also halophiles (microbes that grow in elevated salt concentrations).

To survive and flourish at low temperatures, psychrophiles have to overcome some problems related to permanent cold environments. At low temperatures, enzymes become very rigid, and solute concentrations are at high, perhaps toxic levels [29]. Furthermore, once the water is frozen, ice crystals may pierce the cell membranes, destroying cellular integrity [30].

Membranes of psychrophiles contain increased levels of unsaturated fatty acids that further increase with the reduction in temperature in order to modulate membrane fluidity [31]. Psychrophiles produce cold-adapted enzymes that have high specific activities at low temperatures [32]. These enzymes have the ability to support transcription and translation at low temperatures. Studies have also revealed the presence of certain genes active at low temperature [33]. Moreover, antifreeze proteins have been identified in cold adapted microbes [34]. Such proteins have the ability to bind to ice crystals through a large complementary surface, and hence prevent these crystals from piercing the cell membrane.

2.3. Acidophiles

Acidophiles are microorganisms that grow optimally at pH values of 2.0 [35]. Acidic environments are especially interesting because, in general, the low pH of the habitat is the consequence of microbial metabolism, and not a condition imposed by the system, as is the case for other extreme environments.

Acidophiles oxidize the elemental sulfur (in volcanic areas) or sulfidic minerals (in mine drainage) to obtain energy, which produces extreme acidic environments [36]. Indeed, most of the characterized strict acidophilic microorganisms have been isolated from volcanic areas or acid mine drainage. For example, Archaea *Picrophilus oshimae* and *P. torridus* were isolated from volcanically heated, dry soils in Japan [16]. They thrive at pH 0.7 and 60 °C. These microorganisms maintain the intracellular pH value at 4.6, whereas the other acidophiles maintain their pH at 6.0. *Ferroplasma acidarmanus* was isolated from acid mine drainage in Iron Mountain, California, and is able to grow at a pH of 0 [37]. The Tinto River (Iberian pyritic belt, southwest Spain) is an unusual ecosystem due to its acidity (pH 2.3, buffered by Fe^{3+}), length (100 km), high concentration of heavy metals, and unexpected level of microbial diversity [38]. Due to its size and easy accessibility, the Tinto River is considered an interesting model system for acidic environments.

Environments with low pH condition are a challenge to cellular biochemistry since extreme acidity leads to the denaturation of proteins. Acidophiles protect their proteins by including more amino acids with neutral side-groups and by actively pumping protons out of the cell to maintain constant intracellular pH levels [39].

2.4. Alkaliphiles

Alkaliphiles are microorganisms that grow optimally at pH values above 9.0, often with pH optima around 10.0, while showing little or no growth at near neutral pH values [40]. Alkaline environments may be found in places with high amounts of Ca^{2+} generated by the serpentinization of silicate minerals, as exemplified by the hyperalkaline spring waters seen in Jordan and in the soda lakes and soda deserts of arid and semi-arid areas on Earth, including the high desert in the west of the United States, the East African Rift Valley, and the plateaus of Mongolia, Inner Mongolia, and Tibet [41].

A diversity of microorganisms can live at a pH of 10.5 [42]. Microbial communities live at a pH of 12.9 in the soda lakes of Maqarin, Jordan [43]. Alkaliphiles are often isolated from natural environments that also tend to have high concentrations of NaCl; these are thereby called haloalkalophiles [44].

Under alkaline conditions, the concentrations of hydrogen ions are very low and cells have trouble using ATP-synthase to produce energy and other essential ions, such as magnesium and calcium, which precipitate out of the water as salts (and hence are available only at very low levels) [45]. Base-loving microbes circumvent these problems by actively pumping in these ions and by exporting others to maintain their interior at near-neutrality. Furthermore, the cell wall of alkalophiles acts as a defense barrier from extreme environmental conditions [46].

2.5. Halophiles

Halophiles are microorganisms that grow in elevated salt concentrations, starting from approximately 10% sodium chloride to saturation, and some of them can even survive in salt crystals [47].

The environments where halophilic microorganisms are found include aquatic habitats of varying salinity, salt marshes, surface salt lakes, subterranean salt lakes, and some other places [48]. Two of the largest and best-studied modern hypersaline environments are The Great Salt Lake in Utah and the Dead Sea in the Middle East. Permanently cold hypersaline evaporation ponds are found in dry regions of Antarctica, including Deep Lake, Organic Lake, and Lake Suribati. In these regions, the high salt content may maintain the water liquid at temperatures as low as −20 ℃ [49]. Hypersaline environments are also found within the Earth subsurface, in deep-sea basins associated with undersea salt domes and in subterranean halite deposits from evaporated ancient seas [50].

In response to the salt, all these adapted microorganisms maintain very high concentrations of other solutes in their cytoplasm to keep their insides in osmotic balance with the outside world. Halophilic Archaea keep extremely high concentrations of potassium chloride in their cells [51]. All the proteins in a halophile have to be optimally folded and functional under saturated salt conditions, in much the same way the proteins of hyperthermophiles remain active near 100 °C [52]. Researchers have therefore studied the amino acid sequences, structures, and functional characteristics of halophilic proteins in comparison with thermophilic and mesophilic proteins in order to gain some insights into the evolutionary strategies used to adapt proteins to stress conditions, but the picture remains far from complete [52].

2.6. Piezophiles

Piezophiles are microorganisms that have adapted to high-pressure environments and can grow more easily under high hydrostatic pressure conditions than at atmospheric pressure [53]. Piezophiles are widespread in the seafloor and deep within the Earth's crust. These microbes were isolated from the deepest part of the ocean at a depth of 10.5 Km, and are adapted to pressures of up to 110 Mpa at 2 °C and of 40 Mpa at temperatures above 100 °C [54,55].

In subsurface habitats deep within the Earth's crust, microbes are adapted to high temperatures and extremely limited resources. For example, two different species of thermophilic iron-reducing bacteria were isolated from the granite of Siijan (Sweden) at a depth of 6.7 km [56]. Complex ecosystems have been reported within rocks freshly harvested from 3 km down in South African gold mines [57]. Furthermore, methanogenic microbes have been collected from several hundred meters within basalt rocks in the Columbia River basin (Washington State, USA) [58].

These microbes grow at extremely slow rates and live at low densities. However, considering the vast volume of the upper crust on Earth, this could still constitute a substantial mass of living material. The "deep biota" has been estimated, by some researchers, to exceed the sum total of all surface living systems [59]. In many ways, these subterranean fissures are ideal habitats since they provide a stable ambient with constant flow of chemical energy. Additionally, these environments protect microbes from UV or energetic cosmic radiation, and from the most devastating catastrophes above.

The difficulty in obtaining samples from deep-sea habitats, along with the challenges of conducting biochemical experiments under high pressure conditions in the laboratory, have conspired to make this research field one of the less comprehensively studied. However, recent studies are in progress. Protein-protein interactions are very sensitive to pressure increases, which can be the reason for enzyme dissociation [60]. Pressure is also known to alter gene expression [61]. Furthermore, when pressure increases, or temperature decreases, the molecules in lipid membranes pack tighter, resulting in decreased membrane fluidity [62]. Organisms often circumvent this problem by increasing the proportion of unsaturated fatty acids in their membranes [63].

3. Contributions to Astrobiology

After having examined the diversity of microorganisms and their mechanisms of adaptation to extreme environmental conditions, we now discuss the contribution of these studies to the development of the main themes of astrobiology: the origins of life, the search for extraterrestrial life, and the dispersion of life in the universe.

3.1. Origin of Life

When life appeared on Earth after a period of heavy bombardment, this planet was hotter and more hostile, in comparison with nowadays [64]. The early atmosphere did not contain oxygen until the "great oxidation event", 2.3–2.4 billion years ago. Because life started at least 3.5 billion years ago, it was exclusively anaerobic for at least 1.5 billion years. Heat resistance and anaerobic features are observed in many hyperthermophiles. Thus, the last universal common ancestor (LUCA) of all life on Earth has been suggested to be hyperthermophilic [65]. Indeed, based upon phylogenetic studies, such extremophilic microbes form a cluster on the base of the tree of life [66]. The deepest phylogenetic branches are represented by Aquificales and Thermotogales within bacteria, and Nanoarchaeota, Pyrodictiaceae, and Methanopyraceae within the Archaea [66]. These organisms are found living in hydrothermal vents. For example, the Archaea Strain 121 and *Methanopyrus kandleri*, which are the most hyperthermophilic organisms reported, were isolated from such environments, as well as the living organism with the smallest genome known, the Archaea *Nanoarchaeum equitans* [67].

Hydrothermal vents occur on the seafloor spreading zones and have a global distribution [68]. A hydrothermal vent is a fissure in a planet`s surface from which geothermally heated water emerges. Such vents are commonly found near volcanically active places, areas where tectonic plates are moving apart and where new crust is being formed. The emerging water can approach 350 °C and is prevented from boiling only by the crushing pressure of the deep water. However, the water cools rapidly as it mixes with the frigid ocean around it. The water's carrying capacity drops and many of the dissolved minerals instantly precipitate out. This produces a great cloudy plume of dark particles, which gives these vents their nickname "black smokers". The effluent at black smokers is rich in dissolved transition metals, such as Fe^{2+} and Mn^{2+}, and contains high concentrations of magmatic CO_2, H_2S, and dissolved H_2, with varying amounts of CH_4 [69]. The dissolved gases and metals in black smokers fuel the microbial communities serving as the base of the food chain in these ecosystems [70].

In contrast to the classical Miller experiments to the origins of life, which depend upon the external sources of energy, such as simulated lightning or ultraviolet irradiation, microbes living at hydrothermal vents do not depend on sunlight or oxygen. They grow around the vents and feed upon H_2S and dissolved minerals to produce organic material through the process of chemosynthesis.

However, it is worth to point out that contrasting arguments have been presented to this view. Through comparative studies of the rRNA genes from all domains of life, some authors suggest that the last universal common ancestor (LUCA) evolved in moderate temperatures, and that microbes descending from it became heat-loving only later [71,72].

3.2. The Search for Extraterrestrial Life

One of the prominent goals of astrobiology is to discover life or signs of life on planets beyond Earth. Because currently, there is no direct evidence for life on another planet, our notions of habitability are necessarily constrained by our knowledge of life on Earth. Thus, one question emerges: Are there environments on Earth, which may resemble those we expect to find on other worlds? In this question lies the fundamental connection between the extreme environments on Earth and the search for life on other planets. When we look at our own planet's most challenging environments, we are really looking for clues to what may be the normal conditions on other planets. Therefore, we use our knowledge of the extremes of life on Earth to assess extraterrestrial environments and the plausibility that they can sustain life. Nowadays, based upon the increasing knowledge originated by studies of terrestrial extreme environments, numerous planetary bodies in our Solar System appear to have provided suitable conditions at some point in their history for the emergence of life. Among them, the two candidates that appear more susceptible to sustain life are Mars and Europa.

3.2.1. Mars

Nowadays, the existence of life on the Martian surface seems unlikely, given the extremely desiccating conditions, high UV radiation flux reaching the surface, and the lack of magnetospheric shielding [73]. However, the conditions of the Martian surface early in its history were substantially different from those of today [74,75]. During the first hundreds of millions of years, Mars was probably warmer, presenting an abundance of surface liquid water and a global hydrological cycle [76-78]. Besides, detailed mineral compositions of Mars, obtained by landers and orbiters, have revealed that Mars presented a geochemically active environment [79,80]. The evidence indicates that Mars had an environment similar to Earth, supporting the hypothesis that life may have flourished abundantly on early Mars [81,82]. However, after the first few hundred million years, the environmental histories of these two planets diverged drastically, and today, the conditions of the Martian surface are not suitable for life as we know it [83]. Nevertheless, life could have survived and adapted to the subsurface conditions as microorganisms do in extreme environments on Earth [84]. The most proper terrestrial extreme environments analogous to those of Mars may be the Atacama Desert, the Antarctic Dry Valleys, the Rio Tinto region, and the deep basalt aquifers.

With mean precipitation rates of less than 2 mm a year, the Atacama Desert is one of the driest regions on Earth [85]. Nevertheless, this extreme arid environment hosts relatively abundant communities of endolithic photosynthetic microorganisms within evaporitic crusts of halite [86]. The endolithic environment provides mineral nutrients and protection against harmful radiation for the microbial communities [87]. Previous studies indicated that regions of Mars, such as the Meridiani Planum, once hosted evaporitic environments that resulted in the precipitation of salt-rich deposits [88]. A recently study identified and mapped widespread deposits with a chloride salt component in regions of the southern highlands of Mars [89]. As occurs in the extreme arid region of the Atacama Desert, these deposits could host a variety of microorganisms. Consequently, the Atacama Desert has been considered a potential analog of the extreme arid conditions that dominate the Martian surface [90,91].

The Antarctic Dry Valleys are one of the coldest and driest regions on Earth, since extremely desiccating winds blow continually along the top valleys. During the long bleak winter, the temperature in these dry valleys decreases to −40 °C, reaching 1 °C in summer [92]. Due in part to the hole in the ozone layer, this region has one of the highest levels of UV irradiance on the planet [93]. Life in the Antarctic Dry Valleys is just possible because phototrophic microbes live in the interior of rocks, and are shielded from the hostile environment [94]. Such endolithic habitats are thought to be possible on Mars in the fringe areas of the polar caps, which are almost entirely composed of water ice [95,96]. Recent studies have suggested that the Martian dark dune spots, which are transitional geomorphologic formations in the frost-covered polar regions of Mars, may contain biogenic activity [97].

Rio Tinto is located in the Iberian pyrite belt, southwestern Spain. It is characterized by an extreme acidic environment (mean pH value 2.3) and a high concentration of heavy metals (Fe, Cu, Zn, As, Mn, Cr, *etc.*) [98,99]. These pyrite deposits serve as a habitat to a community supported by chemolithotrophic microbes through oxidation of sulfur and iron [100,101]. Some of these minerals, including hematite and jarosite, have recently been observed in areas of Mars, such as the Meridiani Planum [102]. In fact, Río Tinto's acidic and aqueous conditions produce similar mineralogies to those observed on Mars [103]. Consequently, the subsurface biosphere in the R ó Tinto has been considered a potential biological and geological analog to some areas of Mars [104,105].

Deep basalt aquifers harbor living systems that are absolutely isolated and completely independent of sunlight. These systems are self-reliant in energy and organic substances [106]. These ecosystems, powered by chemoautotrophs, provide the best general terrestrial model for life on Mars. The volcanic minerals in the basaltic rock are rich in highly reduced iron that reacts with water to produce hydrogen, a fuel that the living systems use both to fix carbon and to provide energy from their redox reactions [107]. The products of this redox reaction are water and methane gas; thus, organisms are called methanogens [108]. These anaerobic methanogens are completely independent of oxygen or organic molecules produced by photosynthesis. Basaltic rock is thought to be common in the Martian crust and, since radioactive heating is expected to raise the temperature of the interior, it is believed that a layer of liquid water might exist under the Martian permafrost [109,110].

Recent discoveries demonstrate trace amounts of methane in the Martian atmosphere [111]. The presence of methane on Mars is very intriguing, since it is an unstable gas. Its presence indicates that there must be an active source on the planet in order to maintain such levels in the atmosphere. Mars produces around 270 tonnes of methane annually, but asteroid impacts account for only 0.8% of the total methane production. The presence of substantial amounts of CH_4 would require its recent release from subsurface reservoirs. The lack of current volcanism and hydrothermal activity is not favorable for geologic methane. The source of this CH_4 is uncertain, but it could be either abiotic or biotic (produced by extant life, such as methanogens) [112,113].

3.2.2. Europa

Europa is the sixth moon of the planet, Jupiter, and has a diameter of about 3,121 km. It presents an internal structure similar to the inner terrestrial planets, which consist primarily of silicate rock, and appears to have an iron core [114]. There is a general agreement to the presence of a subsurface water ocean, estimated to be 100 km thick [115], covered with an ice crust estimated to be about 20–30 km thick [116]. Tidal forces and deep-sea vents may provide energy to keep the ice melted [117-119]. Due to the presence of such an ocean, Europa has been considered the most promising target in the search for extant extraterrestrial life within the Solar System. Life could exist within its sub-glacial ocean, perhaps subsisting in an environment similar to Earth's deep-ocean hydrothermal vents or the Antarctic Lake Vostok. Furthermore, the ice layer may provide other potential habits for life.

The young and structurally diverse surface of Europa suggests dynamic activity in its interior [120]. Therefore, the presence of hydrothermal vents is highly probable [121]. These environments are abundant on the floor of the Earth's oceans and are important sources of many elements and organic compounds that are transferred into the hydrosphere [69]. They can sustain life without input from photosynthesis and they sustain fascinating living systems with symbiotic relationships that involve lithoautotrophic microorganisms that use chemical energy to support metazoans [70].

Vostok is a subglacial lake in Antarctica, whose surface is approximately 14 km^2 with a volume of 1.8 km^3 and a maximum depth of 670 m [122]. At Vostok, the ice is 3,750 m thick and has been drilled down to 3,623 m for paleoclimate studies. It is the largest and deepest known subglacial lake and likely the oldest in Antarctica [123]. High pressure, excess gas, low temperature, absence of solar energy, and isolation from surface biota for thousands and perhaps millions of years make this lake a possible analog to Europa's subsurface environment. These factors have stimulated technological projects related to the development of robot exploration systems to be applied in ice satellites [124,125]. The assembly of microbes that was found in the Lake Vostok accretion ice samples indicates that the lake sustains a diverse population of microorganisms and potentially a complex ecosystem, although the concentrations of microbes are expected to be lower than those in most environments on Earth [126].

The Europan surface is an extremely hostile environment, as a result of constant exposure to Jupiter's intense radiation belts [127]. However, this radiation of ions, protons, and electrons does not penetrate ice for more than about a meter and a half. Thus, life may be found on Europa in areas that are protected from the radiation flux, such as in cracks and caves [128]. Also, living systems may live within the ice layer, as microbes do on Earth [129-131]. Furthermore, cracks and fissures that penetrate from the bottom of the ice upwards are likely habitats for life [132].

3.3. Panspermia

The panspermia hypothesis states that the seeds of life exist all over the universe and can be propagated through space from one location to another. For millennia, this idea has been a topic of philosophical debate [133]. However, due to lack of any validation, it remained merely speculative until few decades ago. It is only with the recent discoveries and advances from different fields of research that panspermia has been given serious scientific consideration [134].

This interest was revived in the late 70s by the recognition of Martian meteorites here on Earth [135] which prove beyond doubt that intact rocks can be transferred between the surfaces of planetary bodies in the Solar System. Petrographic analysis of the Martian meteorites and mathematical simulations of impact induced ejection demonstrated that these rocks experienced shocks from 5–10 GPa to 55 GPa [136], heating in the range from 40 °C to 350 °C [137], and acceleration on the order of 3.8×10^6 m/s^2 [138].

Based in this data, mechanisms for the transfer of planetary material have been proposed. The most well accepted mechanism, developed by Melosh [139], indicate that materials can be expelled into interplanetary space under light shocks and modest temperature increases. In fact, recent measurements in the Martian meteorite ALH84001 have shown that it was probably not heated over 40 °C since before it was ejected from Mars [140].

These results led to the question of whether living organisms have been transported between the planets of our Solar System by the same mechanism. The viable transfer from one planet to another requires microorganisms to survive the escape process from one planet, the journey through space, as well as the reentry/impact process on another planet [141]. In this context, a variety of studies has been performed in order to simulate different aspects of lithopanspermia—which postulates that meteors are the transfer vehicles for life through space.

Because of their high resistance to different extreme conditions [142,143], spores of *Bacillus subtilis* are the most widely used model microorganism for these studies [144,145]. However, various other microbes have been used, including vegetative cells of the soil bacteria *Deinococcus radiodurans* and *Rhodococcus erythropolis*, some halophilic Archaea (*Halorubrum* and *Halobacterium* spp.), the cyanobacterium *Chroococcidiopsis*, and others [146,147].

Evidence for the possible interplanetary transfer of biological materials began with experiments testing the resistance of microbes to the space environment [148]. In the space environment, microbes would be subjected to different stresses, including extreme vacuum, desiccation, solar and cosmic radiation, microgravity, and both extreme hot and cold temperatures [149]. Of these factors, solar UV is the most harmful [150].

However, spaceflight experiments demonstrate that with minimal UV shielding, several types of microbes can survive for years at exposures to the harsh environment of space [151]. Furthermore, it is estimated that, if shielded by 2 meters of meteorite, a substantial number of spores would survive after 25 million years in space [152]. On Earth, microbes were brought back to life after 250 million years [153].

Further support to the theory of lithopanspermia has been given by simulation experiments in which model microbes are subjected to ultracentrifugation, hypervelocity, shock pressure, and heating in the range defined for the Martian meteorites found on Earth. These experiments simulate the physical forces that hypothetical endolithic microbes would be subjected to during ejection from one planet and landing upon another. Previous simulation experiments have measured each of these stresses in an isolated manner and the results indicate that spores can survive each stress applied singly [154,155]. The analyses of the combined stresses can be most closely simulated in the laboratory via hypervelocity ballistics experiments. The results demonstrated that microbes could survive rapid acceleration to Mars escape velocities and subsequent impact into surfaces of different compositions [156,157]. Thus, there is a body of evidence suggesting that microbes can survive the conditions of interplanetary transfer from Mars to Earth or from any Mars-like planet to other habitable planets in the same solar system.

The Earth-Mars system is not the only place where natural transfer may occur. The discovery of potentially habitable environments, such as the satellites of Jupiter and Saturn (e.g., Io, Europa, Ganymede, Callisto, Titan, and Enceladus), expands the possibility of interplanetary transfer of life in the Solar System.

Therefore, in recent years, most of the major barriers against the acceptance of panspermia have been demolished and this theory reemerges as a promising field of research.

4. Concluding Remarks

In the last decades, substantial changes have occurred regarding what scientists consider the limits of habitable environmental conditions. Our knowledge about extreme environments and the limits of life on Earth has been greatly expanded. These advances have been fundamental to the development of astrobiology, which studies the origin, evolution, and distribution of life in the universe.

The intense efforts in search of living systems in harsh environments on Earth have given us important clues about the origins of life on Earth. Hydrothermal environments are considered the most proper locals where life may have originated, since these environments provide energy, carbon sources, electron acceptors, and a variety of inorganic surfaces suitable for the formation of biopolymers. For this reason, hydrothermal environments should also be considered the targets for the origins of life in other planets within the Solar System and beyond.

Studies in extreme ambient conditions have changed the previous paradigm that life can only be found on pleasant Earth-like planets. The continuous discovery of living systems thriving in habitats previously thought to be uninhabited has inspired us to speculate more realistically on the forms and constraints that extraterrestrial life may take on other planets. Nowadays, numerous planetary bodies in our Solar System have been suggested to sustain life.

Furthermore, most of the major barriers against the acceptance of panspermia have been demolished when it has been shown that microorganisms can survive the high impact and velocity experienced during the ejection from one planet, the journey through space, and the impact process onto another world.

However, it is worth pointing out that, despite our current advances, we are just beginning to explore and characterize microorganisms living in extreme environments. Most of the deep subterranean and sub-ocean environments remain unknown, and just a few numbers of microbes have been cultured and described. Therefore, for the coming decade, exciting discoveries in the microbial world have the potential to make a great impact on our way of thinking about the nature of life and thereby provide fundamental contributions to astrobiology.

References

1. Horikoshi, K.; Grant, W.D. *Extremophiles: Microbial Life in Extreme Environments*; Wiley-Liss: New York, NY, USA, 1998.
2. Rampelotto, P.H. The search for life on other planets: Sulfur-based, silicon-based, ammonia-based life. *J. Cosmol.* **2009**, *1*, 818-827.
3. Pikuta, E.V.; Hoover, R.B. Microbial extremophiles at the limits of life. *Crit. Rev. Microbiol.* **2007**, *33*, 183-209.
4. Rothschild, L.J.; Mancinelli, R.L. Life in extreme environments. *Nature* **2001**, *409*, 1092-1101.
5. López-García, P. Extremophiles. In *Lectures in Astrobiology*; Gargaud, M., Barbier, B., Martin, H., Reisse, J., Eds.; Springer: New York, NY, USA, 2008.
6. Seckbach, J. *Life as We Know It*; Springer: Dordrecht, The Netherlands, 2006.
7. Westall, F. Early life on Earth: The ancient fossil record. In *Astrobiology*: *Future Perspectives*; Ehrenfreund, P., Irvine, W., Owen, T., Becker, L., Blank, J., Brucato, J., Colangeli, L., Derenne, S., Dutrey, A., Despois, D., Lascano, A., Robert, F., Eds.; Springer: New York, NY, USA, 2005.
8. Cavicchioli, R. Extremophiles and the search for extraterrestrial life. *Astrobiology* **2002**, *2*, 281-292.

9. Horneck, G.; Rettberg, P. *Complete Course in Astrobiology*; Wiley-VCH Verlag GmbH & Co. KGaA: Weinheim, Germany, 2007.
10. Rampelotto, P.H. Are we descendants of extraterrestrials? *J. Cosmol.* **2009**, *1*, 86-88.
11. Madigan, M.T.; Martino, J.M. *Brock Biology of Microorganisms*, 11th ed.; Pearson Education: Upper Saddle River, NJ, USA, 2006.
12. Nakagawa, S.; Takai, K. The isolation of thermophiles from deep-sea hydrothermal environments. In *Methods in Microbiology*: *Extremophiles*; Rainey, F.A., Oren, A., Eds.; Elsevier: New York, NY, USA, 2006.
13. Lund, E.; John, W. Characteristics, development and utilization of geothermal resources. *Q. Bull.* **2007**, *28*, 1-9.
14. Amend, J.P.; Rogers, K.L.; Shock, E.L.; Gurrieri, S.; Inguaggiato, S. Energetics of chemolithoautotrophy in the hydrothermal system of Vulcano Island, southern Italy. *Geobiology* **2003**, *1*, 37-58.
15. Satyanarayana, T.; Raghukumar, C.; Shivaji, S. Extremophilic microbes: Diversity and perspectives. *Curr. Sci.* **2005**, *89*, 78-90.
16. Schleper, C.; Puehler, G.; Holz, I.; Gambacorta, A.; Janekovic, D.; Santarius, U.; Klenk, H.P.; Zillig, W. *Picrophilus* gen. nov. fam. nov.: A novel aerobic, heterotrophic, thermoacidophilic genus and family comprising archaea capable of growth around pH 0. *J. Bacteriol.* **1995**, *177*, 7050-7059.
17. Desbruyères, F.D.; Almeida, A.; Biscoito, M.; Comtet, T.; Khripounoff, A.; Le Bris, N.; Sarradin, P.M.; Segonzac, M. A review of the distribution of hydrothermal vent communities along the northern Mid-Atlantic Ridge: Dispersal *vs.* environmental controls. *Hydrobiologia* **2000**, *440*, 201-216.
18. Daniel, R.M.; Cowan, D.A. Biomolecular stability and life at high temperatures. *Cell. Mol. Life Sci.* **2000**, *57*, 250-264.
19. Takai, K.; Nakamura, K.; Toki, T.; Tsunogai, U.; Miyazaki, M.; Miyazaki, J.; Hirayama, H.; Nakagawa, S.; Nunoura, T.; Horikoshi, K. Cell proliferation at 122 °C and isotopically heavy CH_4 production by a hyperthermophilic methanogen under high-pressure cultivation. *Proc. Nat. Acad. Sci. USA* **2008**, *105*, 10949-10954.
20. Ulrih, N.P.; Gmajner, D.; Raspor, P. Structural and physicochemical properties of polar lipids from thermophilic archaea. *Appl. Microbiol. Biotechnol.* **2009**, *84*, 249-260.
21. Kumar, S.; Nussinov, R. How do thermophilic proteins deal with heat? *Cell. Mol. Life Sci.* **2001**, *58*, 1216-1233.
22. Jaenicke, R. Stability and folding of ultrastable proteins: Eye lens crystallins and enzymes from thermophiles. *FASEB J.* **1996**, *10*, 84-92.
23. Hickey, D.A.; Singer, G.A. Genomic and proteomic adaptations to growth at high temperature. *Genome Biol.* **2004**, *5*, 117.1-117.7.
24. Marguet, E.; Forterre, P. Protection of DNA by salts against thermodegradation at temperatures typical for hyperthermophiles. *Extremophiles* **1998**, *2*, 115-122.
25. Rothschild, L. Extremophiles: Defining the envelope for the search for life in the universe. In *Planetary Systems and the Origins of Life*; Pudritz, R., Higgs, P., Stone, J., Eds.; Cambridge University Press: Cambridge, UK, 2007.

26. Atkins, P.W.; Locke, J.W. *Physical Chemistry*, 7th ed.; Oxford University Press: Oxford, UK, 2004.
27. Margesin, R.; Schinner, F.; Marx, J.C.; Gerday, C. *Psychrophiles*: *From Biodiversity to Biotechnolgy*; Springer: New York, NY, USA, 2008.
28. Rodrigues, D.F.; Jesus, E.C.; Ayala-del-Río, H.L.; Pellizari, V.H.; Gilichinsky, D.; Sepulveda-Torres, L.; Tiedje, J.M. Biogeography of two cold-adapted genera: *Psychrobacter* and *Exiguobacterium*. *ISME J.* **2009**, *3*, 658-665.
29. Cavicchioli, R. Cold-adapted archaea. *Nat. Rev. Microbiol.* **2006**, *4*, 331-343.
30. D'Amico, S.; Collins, T.; Marx, J.C.; Feller, G.; Gerday, C. Psychrophilic microorganisms: Challenges for life. *EMBO J.* **2006**, *7*, 385-389.
31. Nichols, D.; Miller, M.R.; Davies, N.W.; Goodchild, A.; Raftery, M.; Cavicchioli, R. Cold adaptation in the Antarctic archaeon, *Methanococcoides burtonii*, involves membrane lipid unsaturation. *J. Bacteriol.* **2004**, *186*, 8508-8515.
32. Feller, G.; Gerday, C. Psychrophilic enzymes: Hot topics in cold adaptation. *Nat. Rev. Microbiol.* **2003**, *1*, 200-208.
33. Goodchild, A.; Saunders, N.F.W.; Ertan, H.; Raftery, M.; Guilhaus, M.; Curmi, P.M.G.; Cavicchioli, R. A proteomic determination of cold adaptation in the Antarctic archaeon, *Methanococcoides burtonii*. *Mol. Microbiol.* **2004**, *53*, 309-321.
34. Gilbert, J.A.; Hill, P.J.; Dodd, C.E.; Laybourn-Parry, J. Demonstration of antifreeze protein activity in Antarctic lake bacteria. *Microbiology* **2004**, *150*, 171-180.
35. Morozkina, E.V.; Slutskaya, E.S.; Fedorova, T.V.; Tugay, T.I.; Golubeva, L.I.; Koroleva, O.V. Extremophilic microorganisms: Biochemical adaptation and biotechnological application. *Appl. Biochem. Microbiol.* **2010**, *46*, 1-14.
36. Rohwerder, T.; Sand, W. Oxidation of inorganic sulfur compounds in acidophilic prokaryotes. *Eng. Life Sci.* **2007**, *7*, 301-309.
37. Dopson, M.; Baker-Austin, C.; Hind, A.; Bowman, J.P.; Bond, P.L. Characterization of *Ferroplasma* isolates and *Ferroplasma acidarmanus* sp. nov., extreme acidophiles from acid mine drainage and industrial bioleaching environments. *Appl. Environ. Microbiol.* **2004**, *70*, 2079-2088.
38. González-Toril, E.; Llobet-Brossa, E.; Casamayor, E.O.; Amann, R.; Amils, R. Microbial ecology of an extreme acidic environment, the Tinto River. *Appl. Environ. Microbiol.* **2003**, *69*, 4853-4865.
39. Baker-Austin, C.; Dopson, M. Life in acid: pH homeostasis in acidophiles. *Trends Microbiol.* **2007**, *15*, 165-171.
40. Horikoshi, K. Alkaliphiles: Some applications of their products for biotechnology. *Microbiol. Mol. Biol. Rev.* **1999**, *63*, 735-750.
41. Grant, W.D. Alkaline environments and biodiversity. In *Extremophiles*: *Basic Concepts*; Gerdsy, C., Glansdorff, N., Eds.; Encyclopedia of Life Support Systems: Paris, France, 2003.
42. Martins, R.F.; Davids, W.; Al-Sond, W.A.; Levander, F.; Radström, P.; Hatti-Kaul, R. Starch-hydrolyzing bacteria from Ethiopian soda lakes. *Extremophiles* **2001**, *5*, 135-144.

43. Pedersen, K.; Nilsson, E.; Arlinger, J.; Hallbeck, L.; O'Neill, A. Distribution, diversity and activity of microorganisms in the hyper-alkaline spring waters of Maqarin in Jordan. *Extremophiles* **2004**, *8*, 151-164.
44. Gareeb, A.P.; Setati, M.E. Assessment of alkaliphilic haloarchaeal diversity in Sua pan evaporator ponds in Botswana. *Afr. J. Biotechnol.* **2009**, *8*, 259-267.
45. Krulwich, T.A.; Ito, M.; Hicks, D.B.; Gilmour, R.; Guffanti, A.A. pH homeostasis and ATP synthesis: Studies of two processes that necessitate inward proton translocation in extremely alkaliphilic *Bacillus* species. *Extremophiles* **1998**, *2*, 217-222.
46. Horikoshi, K. *Alkaliphiles*: *Genetic Properties and Applications of Enzymes*; Springer: Berlin, Germany, 2006.
47. DasSarma, S.; Arora, P. *Halophiles*, *Encyclopedia of Life Sciences*; Nature Publishing Group: London, UK, 2002.
48. Litchfield, C.D.; Gillevet, P.M. Microbial diversity and complexity in hypersaline environments: A preliminary assessment. *J. Ind. Microbiol. Biotechnol.* **2002**, *28*, 48-55.
49. Madern, D.; Ebel, C.; Zaccai, G. Halophilic adaptation of enzymes. *Extremophiles* **2000**, *4*, 91-98.
50. DasSarma, S. Extreme halophiles are models for Astrobiology. *Microbe* **2006**, *1*, 120-126.
51. Oren, A. Adaptation of halophilic archaea to life at high salt concentrations. In *Salinity*: *Environment—Plants—Molecules*; Lauchli, A., Luttge, U., Eds.; Springer: Dordrecht, The Netherlands, 2004.
52. Michael, T.; Madigan, M.; Orent, A. Thermophilic and halophilic extremophiles. *Curr. Opin. Microbiol.* **1999**, *2*, 265-269.
53. Abe, F.; Horikoshi, K. The biotechnological potential of piezophiles. *Trends Biotechnol.* **2001**, *19*, 102-108.
54. Yayanos, A.A. Microbiology to 10,500 meters in the deep-sea. *Annu. Rev. Microbiol.* **1995**, *49*, 777-805.
55. Kato, C.; Inoue, A.; Horikoshi, K. Isolating and characterizing deep-sea marine microorganisms. *Trends Biotechnol.* **1996**, *14*, 6-12.
56. Kotelnikova, S. Microbial production and oxidation of methane in deep subsurface. *Earth-Sci. Rev.* **2002**, *58*, 367-395.
57. Takai, K.; Moser, D.P.; DeFlaun, M.; Onstott, T.C.; Fredrickson, J.K. Archaeal diversity in waters from deep South African gold mines. *Appl. Environ. Microbiol.* **2001**, *67*, 5750-5760.
58. Thomas-Keprta, K.L.; Wentworth, S.J.; McKay, D.S.; Taunton, A.E.; Allen, C.C.; Romanek, C.S.; Gibson, E.K. Subsurface terrestrial microfossils from Columbia River Basalt samples: Analogs of features in Martian meteorite Allan Hills 84001. *Meteorit. Planet. Sci.* **1997**, *32*, 128-129.
59. Dartnell, L. Extremophiles. In *Life in the Universe*: *A Beginner's Guide*; Dartnell, L., Ed.; Oneworld Publications: Cambridge, UK, 2007.
60. Sharma, A.; Scott, J.H.; Cody, G.D.; Fogel, M.L.; Hazen, R.M.; Hemley, R.J.; Huntress, W.T. Microbial activity at gigapascal pressures. *Science* **2002**, *295*, 1514-1516.
61. Nakasone, K.; Ikegami, A.; Kato, C.; Usami, R.; Horikoshi, K. Mechanisms of gene expression controlled by pressure in deep-sea microorganisms. *Extremophiles* **1998**, *2*, 149-154.

62. Bartlett, D.H. Pressure effects on *in vivo* microbial processes. *Biochim. Biophys. Acta* **2002**, *1595*, 367-381.
63. Aertsen, A.; Meersman, F.; Hendrick, M.E.G.; Vogel, R.F.; Michiels, C.W. Biotechnology under high pressure: Applications and implications. *Trends Biotechnol.* **2009**, *27*, 434-441.
64. Lal, A.K. Origin of life. *Astrophys. Space Sci*. **2008**, *317*, 267-278.
65. Martin, W.; Baross, J.; Kelley, D.; Russell, M.J. Hydrothermal vents and the origin of life. *Nat. Rev. Microbiol.* **2008**, *6*, 805-814.
66. Stetter, K.O. Hyperthermophilic life on Earth—And on Mars? In *Planetary Systems and the Origins of Life*; Pudritz, R., Higgs, P., Stone, J., Eds.; Cambridge University Press: Cambridge, UK, 2007.
67. Waters, E.; Hohn, M.J.; Ahel, I.; Graham, D.E.; Adams, M.D.; Barnstead, M.; Beeson, K.Y.; Bibbs, L.; Bolanos, R.; Keller, M.; Kretz, K.; Lin, X.; Mathur, E.; Ni, J.; Podar, M.; Richardson, T.; Sutton, G.G.; Simon, M.; Soll, D.; Stetter, K.O.; Short, J.M.; Noordewier, M. The genome of *Nanoarchaeum equitans*: Insights into early archaeal evolution and derived parasitism. *Proc. Nat. Acad. Sci. USA* **2003**, *100*, 12984-12988.
68. Baker, E.T.; German, C.R. On the global distribution of hydrothermal vent fields. *Geophys. Monogr.* **2004**, *148*, 245-266.
69. Kelley, D.S.; Baross, J.A.; Delaney, J.R. Volcanoes, fluids, and life at mid-ocean ridge spreading centers. *Annu. Rev. Earth Planet Sci.* **2002**, *30*, 385-491.
70. Takai, K.; Komatsu, T.; Inagaki, F.; Horikoshi, K. Distribution of archaea in a black smoker chimney structure. *Appl. Environ. Microbiol.* **2001**, *67*, 3618-3629.
71. Gribaldo, S.; Brochier-Armanet, C. The origin and evolution of archaea: A state of the art. *Phil. Trans. R. Soc. Lond. B* **2006**, *361*, 1007-1022.
72. Boussau, B.; Blanquart, S.; Necsulea, A.; Lartillot, N.; Gouy, M. Parallel adaptations to high temperatures in the Archaean eon. *Nature* **2008**, *456*, 942-945.
73. Rampelotto, P.H.; Rosa, M.B.; Schuch, N.J.; Schuch, A.P.; Pinheiro, D.K.; Munakata, N. Exobiology at southern Brazil: Spore dosimetry and the UV solar radiation. In *Bioastronomy 2007*: *Molecules*, *Microbes and Extraterrestrial Life*; Werthimer, D., Meech, K., Siefert, J., Mumma, M., Eds.; Astronomical Society of the Pacific: San Francisco, CA, USA, 2010.
74. Krasnopolsky, V.A. Atmospheric chemistry on Venus, Earth, and Mars: Main features and comparison. *Planet. Space Sci.* 2010, (in press).
75. Jakosky, B.M.; Phillips, R.J. Mars' volatile and climate history. *Nature* **2001**, *412,* 237-244.
76. McEwen, A.S.; Hansen, C.J.; Delamere, W.A.; Eliason, E.M.; Herkenhoff, K.E.; Keszthelyi, L.; Gulick, V.C.; Kirk, R.L.; Mellon, M.T.; Grant, J.A.; Thomas, N.; Weitz, C.M.; Squyres, S.W.; Bridges, N.T.; Murchie, S.L.; Seelos, F.; Seelos, K.; Okubo, C.H.; Milazzo, M.P.; Tornabene, L.L.; Jaeger, W.L.; Byrne, S.; Russell, P.S.; Griffes, J.L.; Mart ńez-Alonso, S.; Davatzes, A.; Chuang, F.C.; Thomson, B.J.; Fishbaugh, K.E.; Dundas, C.M.; Kolb, K.J.; Banks, M.E.; Wray, J.J. A closer look at water-related geologic activity on Mars. *Science* **2007**, *317*, 1706-1709.
77. Howard, A.D. Simulating the development of Martian highland landscapes through the interaction of impact cratering, fluvial erosion, and variable hydrologic forcing. *Geomorphology* **2007**, *91*, 332-363.

78. Andrews-Hanna, J.C.; Phillips, R.J.; Zuber, M.T. Meridiani Planum and the global hydrology of Mars. *Science* **2007**, *446*, 7132-7135.
79. Gendrin, A.; Mangold, N.; Bibring, J.P.; Langevin, Y.; Gondet, B.; Poulet, F.; Bonello, G.; Quantin, C.; Mustard, J.; Arvidson, R.; LeMouélic, S. Sulfates in Martian layered terrains: The OMEGA/Mars Express view. *Science* **2005**, *307*, 1587-1591.
80. Bibring, J.P.; Arvidson, R.E.; Gendrin, A.; Gondet, B.; Langevin, Y.; Le Mouelic, S.; Mangold, N.; Morris, R.V.; Mustard, J.F.; Poulet, F.; Quantin, C.; Sotin, C. Coupled ferric oxides and sulfates on the Martian surface. *Science* **2007**, *317*, 1206-1210.
81. Schulze-Makuch, D.; Irwin, L.N.; Lipps, J.H.; LeMone, D.; Dohm, J.M.; Fairén, A.G. Scenarios for the evolution of life on Mars. *J. Geophys. Res*. **2005**, *110*, E12S23.
82. Yung, Y.L.; Russell, M.J.; Parkinson, C.D. The search for life on Mars. *J. Cosmol*. **2010**, *5*, 1121-1130.
83. McKay, C.P.; Andersen, D.T.; Pollard, W.H.; Heldmann, J.L.; Doran, P.T.; Fritsen, C.H.; Priscu, J.C. Polar lakes, streams, and springs as analogs for the hydrological cycle on Mars. In *Water on Mars and Life*; Tokano, T., Ed.; Springer: Berlin, Germany, 2005.
84. Schulze-Makuch, D.; Fairén, A.G.; Davila, A.F. The case for life on Mars. *Int. J. Astrobiology* **2008**, *7*, 117-141.
85. Wierzchos, J.; Ascaso, C.; McKay, C.P. Endolithic cyanobacteria in halite rocks from the hyperarid core of the Atacama Desert. *Astrobiology* **2006**, *6*, 415-422.
86. Warren-Rhodes, K.A.; Rhodes, K.L.; Pointing, S.B.; Ewing, S.A.; Lacap, D.C.; Gómez-Silva, B.; Amundson, R.; Friedmann, E.I.; McKay, C.P. Hypolithic cyanobacteria, dry limit of photosynthesis, and microbial ecology in the hyperarid Atacama Desert. *Microb. Ecol.* **2006**, *52*, 389-398.
87. Davila, A.F.; Gomez-Silva, B.; de los Rios, A.; Ascaso, C.; Olivares, H.; McKay, C.P.; Wierzchos, J. Halite deliquescence facilitates endolithic microbial survival in the hyper—arid core of the Atacama Desert. *J. Geophys. Res. Biogeosci*. **2008**, *113*, doi:10.1029/2007JG000561.
88. Squyres, S.W.; Grotzinger, J.P.; Arvidson, R.E.; Bell, J.F.; Calvin, W.; Christensen, P.R.; Clark, B.C.; Crisp, J.A.; Farrand, W.H.; Herkenhoff, K.E.; Johnson, J.R.; Klingelhöfer, G.; Knoll, A.H.; McLennan, S.M.; McSween, H.Y; Morris, R.V.; Rice, J.W.; Rieder, R.; Soderblom, L.A. *In situ* evidence for an ancient aqueous environment at Meridiani Planum, Mars. *Science* **2004**, *306*, 1709-1714.
89. Osterloo, M.M.; Hamilton, V.E.; Bandfield, J.L.; Glotch, T.D.; Baldridge, A.M.; Christensen, P.R.; Tornabene, L.L.; Anderson, F.S. Chloride-bearing materials in the southern highlands of Mars. *Science* **2008**, *319*, 1651-1654.
90. McKay, C.P.; Friedmann, E.I.; Frankel, R.B.; Bazylinski, D.A. Magnetotactic bacteria on Earth and on Mars. *Astrobiology* **2003**, *2*, 263-270.
91. Navarro-González, R.; Rainey, F.A.; Molina, P.; Bagaley, D.R.; Hollen, B.J.; de la Rosa, J.; Small, A.M.; Quinn, R.C.; Grunthaner, F.J.; Cáceres, L.; Gomez-Silva, B.; McKay, C.P. Mars-like soils in the Atacama Desert, Chile, and the dry limit of microbial life. *Science* **2003**, *7*, 1018-1021.
92. Cary, S.C.; McDonald, I.R.; Barrett, J.E.; Cowan, D.A. On the rocks: The microbiology of Antarctic Dry Valley soils. *Nat. Rev. Microbiol.* **2010**, *8*, 129-138.

93. Cowan, D.; Tow, L.A. Endangered Antarctic environments. *Annu. Rev. Microbiol.* **2004**, *58*, 649-690.
94. Pointing, S.B.; Chan, Y.; Lacap, D.C.; Lau, M.C.; Jurgens, J.A.; Farrell, R.L. Highly specialized microbial diversity in hyper-arid polar desert. *Proc. Nat. Acad. Sci. USA* **2009**, *106*, 19964-19969.
95. Heldmann, J.L.; Toon, O.B.; Pollard, W.H.; Mellon, M.T.; Pitlick, J.; McKay, C.P.; Andersen, D.T. Formation of Martian gullies by the action of liquid water flowing under current Martian environmental conditions. *J. Geophys. Res.* **2005**, *110*, doi:10.1029/2004JE002261.
96. Malin, M.C.; Edgett, K.S.; Posiolova, L.V.; McColley, S.M.; Noe Dobrea, E.Z. Present-day impact cratering rate and contemporary gully activity on Mars. *Science* **2006**, *314*, 1573-1577.
97. Szathmary, E.; Ganti, T.; Pocs, T.; Horvath, A.; Kereszturi, A.; Berczi, S.; Sik, A. Life in the Dark Dune Spots of Mars: A Testable Hypothesis. In *Planetary Systems and the Origins of Life*; Pudritz, R., Higgs, P., Stone, J., Eds.; Cambridge University Press: Cambridge, UK, 2007.
98. Fernández-Remolar, D.C.; Morris, R.V.; Gruener, J.E.; Amils, R.; Knoll, A.H. The Río Tinto basin, Spain: Mineralogy, sedimentary geobiology, and implications for interpretation of outcrop rocks at Meridiani Planum, Mars. *Earth Planet. Sci. Lett.* **2005**, *240*, 149-167.
99. Prieto-Ballesteros, O.; Martínez-Frías, J.; Schutt, J.; Sutter, B.; Heldmann, J.L.; Bell, M.S.; Battler, M.; Cannon, H.; Gómez-Elvira, J.; Stoker, C.R. The subsurface geology of Río Tinto: Material examined during a simulated Mars Drilling Mission for the Mars Astrobiology Research and Technology Experiment (MARTE). *Astrobiology* **2008**, *8*, 1013-1021.
100. Stoker, C.R.; Cannon, H.N.; Dunagan, S.E.; Lemke, L.G.; Glass, B.J.; Miller, D.; Gomez-Elvira, J.; Davis, K.; Zavaleta, J.; Winterholler, A.; Roman, M.; Rodriguez-Manfredi, J.A.; Bonaccorsi, R.; Bell, M.S.; Brown, A.; Battler, M.; Chen, B.; Cooper, G.; Davidson, M.; Fernández-Remolar, D.; Gonzales-Pastor, E.; Heldmann, J.L.; Martínez-Frías, J.; Parro, V.; Prieto-Ballesteros, O.; Sutter, B.; Schuerger, A.C.; Schutt, J.; Rull, F. The 2005 MARTE robotic drilling experiment in Río Tinto, Spain: Objectives, approach, and results of a simulated mission to search for life in the Martian subsurface. *Astrobiology* **2008**, *8*, 921-945.
101. Fernández-Remolar, D.C.; Prieto-Ballesteros, O.; Rodríguez, N.; Gómez, F.; Amils, R.; Gómez-Elvira, J.; Stoker, C.R. Underground habitats in the Río Tinto Basin: A model for subsurface life habitats on Mars. *Astrobiology* **2008**, *8*, 1023-1047.
102. Christensen, P.R.; Wyatt, M.B.; Glotch, T.D.; Rogers, A.D.; Anwar, S.; Arvidson, R.E.; Bandfield, J.L.; Blaney, D.L.; Budney, C.; Calvin, W.M.; Fallacaro, A.; Fergason, R.L.; Gorelick, N.; Graff, T.G.; Hamilton, V.E.; Hayes, A.G.; Johnson, J.R.; Knudson, A.T.; McSween, H.Y.; Mehall, G.L.; Mehall, L.K.; Moersch, J.E.; Morris, R.V.; Smith, M.D.; Squyres, S.W.; Ruff, S.W.; Wolff, M.J. Mineralogy at Meridiani Planum from the Mini-TES experiment on the Opportunity Rover. *Science* **2004**, *306*, 1733-1739.
103. Klingelhöfer, G.; Morris, R.V.; Bernhardt, B.; Schroder, C.; Rodionov, D.S.; de Souza, P.A.; Yen, A.; Gellert, R.; Evlanov, E.N.; Zubkov, B.; Foh, J.; Bonnes, U.; Kankeleit, E.; Gutlich, P.; Ming, D.W.; Renz, F.; Wdowiak, T.; Squyres, S.W.; Arvidson, R.E. Jarosite and hematite at Meridiani Planum from Opportunity's Mössbauer spectrometer. *Science* **2004**, *306*, 1740-1745.

104. Amils, R.; González-Toril, E.; Fernández-Remolar, D.C.; Gómez, F.; Aguilera, A.; Rodríguez, N.; Malki, M.; García-Moyano, A.; Fairén, A.G.; de la Fuente, V.; Sanz, J.L. Extreme environments as Mars terrestrial analogs: The Río Tinto case. *Planet. Space Sci.* **2007**, *55*, 370-381.
105. Bonaccorsi, R.; Stoker, C.R. Science results from a Mars drilling simulation (Río Tinto, Spain) and ground truth for remote science observations. *Astrobiology* **2008**, *8*, 967-985.
106. Stevens, T.O.; McKinley, J.P. Lithoautotrophic microbial ecosystems in deep basalt aquifers. *Science* **1995**, *270*, 450-455.
107. Krumholz, L.R. Microbial communities in the deep subsurface. *Hydrogeol. J.* **2000**, *8*, 4-10.
108. Chapelle, F.H.; O'Neill, K.; Bradley, P.M.; Methe, B.A.; Ciufo, S.A.; Knobel, L.L.; Lovley, D.R. A hydrogen-based subsurface microbial community dominated by methanogens. *Nature* **2002**, *415*, 312-315.
109. Boynton, W.V.; Feldman, W.C.; Squyres, S.W.; Prettyman, T.H.; Bruckner, J.; Evans, L.G.; Reedy, R.C.; Starr, R.; Arnold, J.R.; Drake, D.M.; Englert, P.A.; Metzger, A.E.; Mitrofanov, I.; Trombka, J.I.; D'Uston, C.; Wanke, H.; Gasnault, O.; Hamara, D.K.; Janes, D.M.; Marcialis, R.L.; Maurice, S.; Mikheeva, I.; Taylor, G.J.; Tokar, R.; Shinohara, C. Distribution of hydrogen in the near surface of Mars: Evidence for subsurface ice deposits. *Science* **2002**, *297*, 81-85.
110. Plaut, J.J.; Picardi, G.; Safaeinili, A.; Ivanov,A.B.; Milkovich, S.M.; Cicchetti, A.; Kofman, W.; Mouginot, J.; Farrell, W.M.; Phillips, R.J.; Clifford, S.M.; Frigeri, A.; Orosei, R.; Federico, C.; Williams, I.P.; Gurnett, D.A.; Nielsen, E.; Hagfors, T.; Heggy, E.; Stofan, E.R.; Plettemeier, D.; Watters, T.R.; Leuschen, C.J.; Edenhofer, P. Subsurface radar sounding of the South Pole deposits of Mars. *Science* **2007**, *316*, 92-95.
111. Formisano, V.; Atreya, S.; Encrenaz, T.; Ignatiev, N.; Giuranna, M. Detection of methane in the atmosphere of Mars. *Science* **2004**, *306*, 1758-1761.
112. Atreya, S.K.; Mahaffy, P.R.; Wong, A.S. Methane and related trace species on Mars: Origin, loss, implications for life, and habitability. *Planet. Space Sci*. **2007**, *55*, 358-369.
113. Onstott, T.C.; McGown, D.; Kessler, J.; Lollar, B.S.; Lehmann, K.K. Clifford, S.M. Martian CH4: Sources, flux, and detection. *Astrobiology* **2006**, *6*, 377-395.
114. Kuskov, O.L.; Kronrod, V.A. Internal structure of Europa and Callisto. *Icarus* **2005**, *177*, 550-556.
115. Greenberg, R.; Geissler, P.; Hoppa, G.; Tufts, B.R. Tidal-tectonic processing and their implications for the character of Europa's icy crust. *Rev. Geophys*. **2002**, *40*, 1-33.
116. Ruiz, J.; Alvarez-Gómez, J.A.; Tejero, R.; Sánchez, N. Heat flow and thickness of a convective ice shell on Europa for grain size—dependent rheologies. *Icarus* **2007**, *190*, 145-154.
117. Marion, G.M.; Fritsen, C.H.; Eicken, H.; Payne, M.C. The search for life on Europa: Limiting environmental factors, potential habitats, and Earth analogues. Astrobiology 2003, *3*, 785-811.
118. Greenberg, R. Tides and the biosphere of Europa. *Am. Sci.* **2002**, *90*, 48-55.
119. Lipps, J.H.; Rieboldt, S. Habitats and taphonomy of Europa. *Icarus* **2005**, *177*, 515-527.
120. Marcos, R.F.; Nissar, A. Possible detection of volcanic activity on Europa: Analysis of an optical transient event. *Earth Moon Planets* **2000**, *88*, 167-175.

121. McCollom, T.M. Methanogenesis as a potential source of chemical energy for primary biomass production by autotrophic organisms in hydrothermal systems on Europa. *J. Geophys. Res.* **1999**, *104*, 30729-30742.
122. Priscu, J.C.; Adams, E.E.; Lyons, W.B.; Voytek, M.A.; Mogk, D.W.; Brown, R.L.; McKay, C.P.; Takacs, C.D.; Welch, K.A.; Wolf, C.F.; Kirshtein, J.D.; Avci, R. Geomicrobiology of subglacial ice above Lake Vostok, Antarctica. *Science* **1999**, *286*, 2141-2144.
123. Siegert, M.J.; Ellis-Evans, J.C.; Tranter, M.; Mayer, C.; Petit, J.; Salamatin, A.; Priscu, J.C. Physical, chemical, and biological processes in Lake Vostok and other Antarctic subglacial lakes. *Nature* **2001**, *414*, 603-609.
124. Salamatin, A.N.; Tsyganova, E.; Lipenkov, V.; Petit, J.R. Vostok (Antarctica) ice-core time-scale from datings of different origins. *Ann. Glaciol*. **2004**, *39*, 283-292.
125. Rogers, S.O.; Theraisnathan, V.; Ma, L.J.; Zhao, Y.; Zhang, G.; Shin, S.G.; Castello, J.D.; Starmer, W.T. Comparisons of protocols to decontaminate environmental ice samples for biological and molecular examinations. *Appl. Environ. Microbiol*. **2004**, *70*, 2540-2544.
126. D'Elia, T.; Veerapaneni, R.; Rogers, S.O. Isolation of microbes from Lake Vostok accretion ice. *Appl. Environ. Microbiol.* **2008**, *74*, 4962-4965.
127. Paranicas, C.; Mauk, B.H.; Khurana, K.; Jun, I.; Garrett, H.; Krupp, N.; Roussos, E. Europa's nearsurface radiation environment. *Geophys. Res. Lett.* **2007**, *34*, doi:10.1029/2007GL030834.
128. Figueredo, P.H.; Greeley, R.; Neuer, S.; Irwin, L.; Schulze-Makuch, D. Locating potential biosignatures on Europa from surface geology observations. *Astrobiology* **2003**, *3*, 851-861.
129. Gilichinsky, D.A. Permafrost model of extraterrestrial habitat. In *Astrobiology*: *The Quest for the Conditions of Life*; Horneck, G., Baumstark-Khan, C., Eds.; Springer: Berlin, Germany, 2002.
130. Gilichinsky, D.A.; Rivkina, E.; Bakermans, C.; Shcherbakova, V.; Petrovskaya, L.; Ozerskaya, S.; Ivanushkina, N.; Kochkina, G.; Laurinavichuis, K.; Pecheritsina, S.; Fattakhova, R.; Tiedje, J.M. Biodiversity of cryopegs in permafrost. *FEMS Microbiol. Ecol.* **2005**, *53*, 117-128.
131. Thomas, D.N.; Dieckmann, G.S. Antarctic Sea ice—a habitat for extremophiles. *Science* **2002**, *295*, 641-644.
132. Greenberg. R. Europa, the ocean moon: Tides, permeable ice, and life. In *Planetary Systems and the Origins of Life*; Pudritz, R., Higgs, P., Stone, J., Eds.; Cambridge University Press: Cambridge, UK, 2007.
133. Raulin-Cerceau, F.; Maurel, M.C.; Schneider, J. From Panspermia to Bioastronomy: The evolution of the hypothesis of universal life. *Orig. Life. Evol. Biosph.* **1998**, *28*, 597-612.
134. Rampelotto, P.H. Are we descendants of extraterrestrials? *J. Cosmol.* **2009**, *1*, 86-88.
135. Fritz J.; Artemieva N.; Greshake, A. Ejection of Martian meteorites. *Meteoritics Planet. Sci.* **2005**, *40*, 1393-1411.
136. Nyquist, L.E.; Bogard, D.D.; Shih, C.Y.; Greshake, A.; Stöffler, D.; Eugster, O. Ages and geologic histories of Martian meteorites. *Space Sci. Rev*. **2001**, *96*, 105-164.
137. Shuster, D.L.; Weiss, B.P. Martian surface paleotemperatures from thermochronology of meteorites. *Science* **2005**, *309*, 594-597.
138. Mastrapa, R.M.E.; Glanzberg, H.; Head, J.N.; Melosh, H.J.; Nicholson, W.L. Survival of bacteria exposed to extreme acceleration: Implications for panspermia. *Earth Planet. Sci. Lett.* **2001**, *189*, 1-8.

139. Melosh, H.J. The rocky road to panspermia. *Nature* **1988**, *21*, 687-688.
140. Weiss, B.P.; Kirschvink, J.L.; Baudenbacher, F.J.; Vali, H.; Peters, N.T.; MacDonald, F.A.; Wikswo, J.P. A low temperature transfer of ALH84001 from Mars to Earth. *Science* **2000**, *290*, 791-795.
141. Horneck, G.; Stoeffler, D.; Eschweiler, U.; Hornemann, U. Bacterial spores survive simulated meteorite impact. *Icarus* **2001**, *149*, 285-290.
142. Rosa, M.B.; Rampelotto, P.H.; Schuch, N.J.; Schuch, A.P.; Munakata, N. Spore dosimetry: *Bacillus subtilis* TKJ6312 as biosensor of biologically effective solar radiation. *Quim. Nova* **2009**, *32*, 282-285.
143. Hecker, M.; Völker, U. Non-specific, general and multiple stress resistance of growth-restricted *Bacillus subtilis* cells by the expression of the σB regulon. *Mol. Microbiol.* **1998**, *29*, 1129-1136.
144. Rampelotto, P.H.; Rosa, M.B.; Schuch, A.P.; Pinheiro, D.K.; Schuch, N.J.; Munakata, N. Exobiology research in the south of Brazil aiming the monitoring of the biogenically-effective solar radiation. *Astrobiology* **2007**, *7*, 502-540.
145. Rampelotto, P.H.; Rosa, M.B.; Schuch, N.J.; Munakata, N. Exobiological application of spore dosimeter in studies involving solar UV radiation. *Orig. Life. Evol. Biosph.* **2009**, *39*, 373-374.
146. De Vera, J.P.; Horneck, G.; Rettberg, P.; Ott, S. The potential of the lichen symbiosis to cope with extreme conditions of outer space—I. Influence of UV radiation and space vacuum on the vitality of lichen symbiosis and germination capacity. *Intern. J. Astrobiol*. **2003**, *1*, 285-293.
147. Nicholson, W.L. Ancient micronauts: Interplanetary transport of microbes by cosmic impacts. *Trends. Microbiol.* **2009**, *17*, 243-250.
148. Horneck, G. Responses of *Bacillus subtilis* spores to space environment: Results from experiments in space. *Origin. Life Evol. Biosph.* **1993**, *23*, 37-52.
149. Nicholson, W.L.; Munakata, N.; Horneck, G.; Melosh, H.J.; Setlow, P. Resistance of *Bacillus* endospores to extreme terrestrial and extraterrestrial environments. *Microbiol. Mol. Biol. Rev.* **2000**, *64*, 548-572.
150. Horneck, G.; Buecker, H.; Reitz, G. Long-term survival of bacterial spores in space. *Adv. Space Res.* **1994**, *10*, 41-45.
151. Rettberg, P.; Eschweiler, U.; Strauch, K.; Reitz, G.; Horneck, G.; Wänke, H.; Brack, A.; Barbier, B. Survival of microorganisms in space protected by meteorite material: Results of the experiment "EXOBIOLOGIE" of the PERSEUS mission. *Adv. Space Res.* **2002**, *30*, 1539-1545.
152. Horneck, G.; Miliekowsky, C.; Melosh, H.J.; Wilson, J.W.; Cucinotta, F.A.; Gladman, B. Viable transfer of microorganisms in the solar system and beyond. In *Astrobiology*: *The Quest for the Conditions of Life*; Horneck, G., Baumstark-Khan, C., Eds.; Springer: Berlin, Germany, 2002.
153. Vreeland, R.H.; Rosenzweig, W.D.; Powers, D.W. Isolation of a 250 million year-old halotolerant bacterium from a primary salt crystal. *Nature* **2000**, *407*, 897-900.
154. Stoffler, D.; Horneck, G.; Ott, S.; Hornemann, U.; Cockell, C.S.; Moeller, R.; Meyer, C.; de Vera, J.P.; Fritz, J.; Artemieva, N.A. Experimental evidence for the potential impact ejection of viable microorganisms from Mars and Mars-like planets. *Icarus* **2007**, *186*, 585-588.
155. Burchell, M.J.; Mann, J.R.; Bunch, A.W. Survival of bacteria and spores under extreme shock pressures. *Mon. Not. R. Astron. Soc.* **2004**, *352*, 1273-1278.

156. Horneck, G.; Stoffler, D.; Ott, S.; Hornemann, U.; Cockell, C.S.; Moeller, R.; Meyer, C.; de Vera, J.P.; Fritz, J.; Schade, S.; Artemieva, N.A. Microbial rock inhabitants survive hypervelocity impacts on Mars-like host planets: First phase of Lithopanspermia experimentally tested. *Astrobiology* **2008**, *8*, 17-44.
157. Fajardo-Cavazos, P.; Langenhorst, F.; Melosh, H.J.; Nicholson, W.L. Bacterial spores in granite survive hypervelocity launch by spallation: Implications for Lithopanspermia. *Astrobiology* **2009**, *9*, 647-657.