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VISUALIZATION OF SOIL MICROORGANISMS IN EXTREME ENVIRONMENTS BY FLUORESCENT MICROSCOPY
Theme 1: Origin of Biosphere and evolution of geo-biological systems

ANCIENT WEATHERING CRUSTS AND BIOTIC COLONIZATION OF LAND

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There are a lot of bacterial paleontological achievements that was made last years. But most of them dealt with the microfossils found in marine sediments, i.e. studied ancient microbes lived in marine environments. So a wrong opinion, that life in Archaean – Early Proterozoic times was restricted by marine conditions, could appear. But there are strong evidences concerning life, existed on land, i.e. on parts of Earth’s surface without basin reservoirs. These evidences of microbial land colonization are ancient weathering crusts and microfossil remains found in them. Weathering products – sedimentary rocks – are fixed already for the very early stages of the Earth evolution. The presence of microbial mats on the land surface as early as 2.7-2.6 GA was affirmed by Y. Watanabe, J.E.J. Martini and H. Ohmoto in 2000. But the fossilized remains of AR-PR1 land microorganisms have not been discovered till 2008 by A.Yu. Rozanov and M.M. Astafieva. The objects of our researches were different the most ancient weathering crusts of Karelia and Kola peninsular with the age from 2.8 GA. As a result of our investigations rather morphologically diverse and rich complex of fossilized microbial forms was found. Filament or thread-like forms played leading role, cocci and oval forms were subordinated. There also biofilms and large (diameter < 10 µ) spheres. Sometimes the whole fragments of crusts were consisted from microbial remains (destroyed cocci, filament shreds, dump-bell forms etc.). So, we can say that beginning from Archaean microorganisms – bacteria, probably, cyanobacteria and, perhaps, even eucaryots accompanied and promote formation of weathering crusts. So it is impossible to speak about pure chemical nature of all the most ancient weathering crusts, biological component was presented in their formation even in those times. And the most important conclusion – the land was colonized by microorganisms may be from the very appearance of sedimentary rocks. Due to the fact, that the most ancient Earth rocks (AR-PR1) considered to be the model objects for studying rocks, comprising astromaterials, our investigations are of great value for astrobiology also.

The study was executed within the framework of the complex program of basic research of the Presidium of the Russian Academy of Sciences “Origin of Biosphere and Evolution of Geo-biological Systems” (subprogram II) and was supported by the Russian Foundation for Basic Research, projects 08-04-00484 and SS-4207.2008.5.

EMERGENCE OF THE CHEMOTROPHIC METABOLISM SYSTEMS IN THE DEEP

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A simplest metabolic system, a combined autocatalytic 3-hydroxypropionate (3-HP) and reductive citrate (RC) cycles, was submitted. This bicycle contains a succinate – fumarate core, capable of the electron flow switch in forward or reverse direction depending on redox potential of the geochemical environment. Competition between this autocatalytic cycles in the conditions of the ancient Earth led to physicochemical selection, major factors of which were temperature, redox potential and mineral composition of the surrounding hydrothermal environment. The correspondence of cycles to these conditions was criterion of the selection. At reconstruction of the first "minimal" cell the determinative role of the metabolism is revealed in the chemoton model [1, 2], which represents three stoichiometrically coupled autocatalytic subsystems: a metabolic bicycle, a system of template replication and a membrane surrounding them. Autocatalytic bicycle operates inside of a similar protocell with the formation not only itself metabolites, but also building elements for a membrane and a template synthesis. At steady state inflow of hydrogen and hydrocarbons from the hydrothermal pool [3] there is a possibility of functioning of a protocell bicycle, as a chemoheterotrophic metabolic system. The natural selection under influence of ancient hydrothermal environment resulted in the divergence of protocells with formation of ancestral phyla Aquificae and Chloroflexi, the extant species of which function on the basis of RC and 3-HP cycles, respectively.

References:
catalyst, heat time, amino acids ratio). It was shown that the number of various structures into the synthesized products mixtures was much smaller compared to the statistically possible value. This study was partly supported by Program P-15 of Presidium of Russian Academy of Sciences.

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USING EXTREME SOLAR EVENTS AS PROXY FOR THE ACTIVE YOUNG SUN: IMPLICATIONS FOR PLANETARY ATMOSPHERE EVOLUTION

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The CHAMP satellite is a low Earth orbiting (LEO) mission which carries among other instruments a high-precision accelerometer on board which is used in this study to investigate the temporal and spatial variation of the atmospheric density caused by the so-called Halloween (extreme solar activity events) events at an altitude of about 400 km. We focus on the variation of the neutral atmospheric density and related temperature in the upper thermosphere during the extreme solar events and compare the absolute density values with state of the art density models such as NRLMISE-00 and Jacchia-Bowman 2008. These atmospheric disturbances originated from CMEs associated with a solar flare of magnitude X17.2 and caused density enhancements up to about 300 - 400% which may lead to significant local rise in exobase temperatures compared to quiet solar conditions. An analysis of these events is used for the investigation of the connection between such extreme solar events and the activity of solar proxies with different age. Solar data of the Halloween event in Oct./Nov. 2003 are used as proxy for the active young sun. Therefore, the data sets especially those of the 28th October 2003 are retrieved from the Coronal Diagnostic Spectrometer (CDS) onboard SOHO. The detected flare is measured in the extreme Ultraviolet at about 300 - 380 Angstroms and 510 - 630 Angstroms, respectively, which is an important range for studying the solar influence on upper atmospheres. After applying correction and calibration procedures to the data they are compared to the quiet sun and afterwards to spectra of young solar-like stars of different age. Solar flare spectra can simulate the situation of the young sun if they match spectra of young solar analogues. We show that this approach is important for studies which are related to the evolution of the early nitrogen-rich atmosphere of Earth and for Earth-like nitrogen atmospheres of exoplanets orbiting around active host stars.

EVIDENCE OF DIVERSE SUBTERRESTIAL ECOSYSTEMS ~ 1000 MA AGO

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New fossil remains have been discovered from the well-known organic-walled Lakhanda microbiota (1015–1025 Ma, Uchur-Maya Region, Southeastern Siberia) show that there exists advanced prokaryote-eukaryote community. Ancient organisms recorded in the mats and biofilms of different architectures. The microfossils association has a structure close to the observed modern communities, and includes well-known genera of fossil filamentous blue-green algae that are attributable to the morphology of genus Oscillatoriopsis, Palaeolyngbya, Polytrichoides, Siphonophycus and Ostiana. Bacterial organisms, played an important geochemical role in the community, recognized by the presence of narrow blank covers, rod and coccoid cells. The microfossils have characters observed in microscopic fungi, such as a reticulate mycelium, formed by anastomizing hyphae of non-cellular structure and of variable width. The presence on hyphae of knoblike trapping structures and secretion of adhesive enzymes suggest a similarity with modern nematophagous fungi. Microorganisms associated with the adhesive hyphae consist of colonial green unicellular algae and thin bacterial sheaths, which may indicate mutually beneficial relationships between morphologically and biologically different partners. On the border of the Meso-Neoproterozoic in the atmosphere was enough oxygen for the emergence of new levels of eukaryotes organizational complexity. At this point finds a large, morphologically very diverse group of abundant siphonophycean green algae and complicated structure of two-layer tubular organisms with the attachment disk (Eosolenides), who first detected the signs endosymbiosis of unicellular algae (German, Podkovyrov, 2009). The biota inhabited a warm, shallow, epicontinental basin rich in nutrients into which were dispersed clay minerals, from weathering of continental crust. Transgressive-regressive phenomena in the Lakhanda basin led to the formation of local shoals. Such sites could hypothetically get used lakhandinian benthic community, including a symbiotic association of cyanobacteria and fungi, capable of binding (glue) clay particles absorb bottom of the reservoir and adapted to the subterrestrial mode of existence. Detected in the biofilms (German, 1979), the benthic community to complement the green xanthophytic Palaeovaucheria, sessile forms Cypandinia, organisms with basal discs Eosolenides and nematode-like Rugosoopsis. Alleged nematode nature of these organisms gives additional arguments in the presence of microbiota another group of microorganisms, which are considered one of the first potential candidates in the development of land biocenoses. Such communities is dominated by associations of fungi, algae, cyanobacteria and heterotrophic bacteria are usual for recent “Subaerial biofilm” (SAB, Gorbushina, 2009) communities that develop on solid surfaces exposed to the atmosphere. The ~ 1 Ga Lakhanda microbiota was able to at least partially to colonize physically unstable sedimentary environments, and to act as successful pioneers in the biostabilization process.

**3D-VISUALIZATION OF WELL-PRESERVED SUBSURFACE CARBONACEOUS FILAMENTS OF ARCHAEAN AGE FROM THE PILBARA CRATON, AUSTRALIA**

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Banded hydrothermal quartz-chalcedony filling former solution cavities in the ~3430 Ma Strelley Pool Chert, Pilbara Craton, Australia (Trendall locality), was formed by encrustation of a filamentous substrate. These features were formerly interpreted as stalactites [1]. They also occur in the slightly older Duffer formation (3467 Ma). After recognition of many similar Phanerozoic samples of likely microbial origin [2], we investigated Archaean samples in a new light [3]. While poorly preserved filaments are common, we now have also identified well-preserved filaments forming web-like fabrics in the central part of some of the quartz precipitates. Raman-spectroscopy demonstrates that the carbonaceous material present in central filaments and different bands of younger laminated quartz precipitates is thermally altered (~340 °C) and thus indigenous and very different from recent organic contaminations on the surface of the rock (dominated by carotene). 3D-visualization of the
filamentous fabric was performed by optical microscopy of a doubly polished thick section. Stacks of 45-100 images were taken of areas containing well-preserved filaments. Using Automontage software, image stacks were combined into single images displaying the in-focus areas of each image of the stack. For 3-D visualization, red-blue anaglyphs were produced from the same stacks. Automontage images and anaglyphs clearly show the filamentous nature of the centre of the quartz precipitates. Based on these images, a web of carbonaceous filaments with a diameter of ~0.5 μm was the basis of quartz precipitation. The preservation of filaments within the same sample is highly variable. Apparent filament diameters of up to 20 μm in less well-preserved areas demonstrate that taphonomic processes affected the sample in a heterogeneous way. We interpret the filaments as remnants of a subsurface microbiota in cavities within the Strelley Pool chert. The arrangement of “stalactites” perpendicular to bedding demonstrated a formation before regional tilting of the strata occurred at 3300-2950 Ma [4]. This limits the age of the filamentous fabrics to the Archaean and is evidence for the presence of early subsurface life and of taphonomic processes in the Archaean that yield macroscopic features from encrustation of microscopic filaments.

References:

REDUCTION OF OXIDIZED NITROGEN COMPOUNDS DURING SERPENTINIZATION OF MAFIC ROCKS AND PREBIOTIC FORMATION OF EARLY LIFE MOLECULES
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Hydrogen cyanide is an excellent organic reagent and is central to most of the reaction pathways leading to abiotic formation of simple organic compounds containing nitrogen, such as amino acids, purines and pyrimidines. Reduced carbon and nitrogen precursor compounds for the synthesis of HCN may be formed under off-axis hydrothermal conditions in oceanic lithosphere in the presence of native Fe and Ni and are adsorbed on authigenic layer silicates and zeolites. The native metals as well as the molecular hydrogen reducing CO2 to CO/CH4 and NO3-/NO2- to NH3/NH4+ are a result of serpentinization of mafic rocks. Oceanic plates are conveyor belts of reduced carbon and nitrogen compounds from the off-axis hydrothermal environments to the subduction zones, where compaction, dehydration, desiccation and diagenetic reactions affect the organic precursors [1]. CO/CH4 and NH3/NH4+ in fluids distilled out of layer silicates and zeolites in the subducting plate at an early stage of subduction will react upon heating and form HCN, which is then available for further organic reactions to, for instance, carbohydrates, nucleosides or even nucleotides due to phosphorylation by pyrophosphate under the alkaline conditions prevalent in hydrated mantle rocks of the overriding plate [2]. Convergent margins in the initial phase of subduction must, therefore, be considered the most potent sites for prebiotic organic reactions on Earth. This means that processes leading to the origin of life probably are possible only on planets where some kind of plate tectonics occurs.
The problem of homochirality is of crucial importance for the origin of life. An extraterrestrial origin requires that biomolecules pass through space environments where strong radiations or/and high temperatures are present. This naturally implies the possibility of degradation of organic molecules and losing of their optical activity, i.e. racemization. The space bodies have not the same thermal history and the survivability of the molecules delivered to the earth can be influenced by different factors, one of them is the chemical composition of the space body. Indeed some of its components such minerals can act as catalysts for the molecule decomposition and racemization. The aim of the present work is to study the influence of temperature and influence of different minerals like silicates, ilmenite, wuestite, Mg2Si, etc. on the decomposition and the racemization of the most simple chiral aromatic molecule, i.e. (R)- or (S)-1-phenylethanol. In addition, other benzylic alcohols have also been studied to compare their behaviour with 1-phenylethanol. A Py-GC-MS technique was used to investigate this phenomenon. The minerals are deposited directly on the quartz liner of the pyrolizer. The gaschromatograph is equipped with a chiral column to discriminate enantiomers and diastereoisomers. A quadrupole mass spectrometer acts as the gas chromatograph detector. Various pyrolytic experiments, at temperatures between 100 and 700°C with and without minerals, were performed. Obtained results demonstrate that the rate of pyrolysis and racemization of 1-phenylethanol depends on the presence of the catalyst and on its nature. In the absence of catalyst, pure (R)- or (S)-1-phenylethanol enantiomers show poor sign of racemization up to 500°C. However, when the pyrolysis is carried out on minerals the enantiomeric excess never has been found lower than 75% in the range of temperature applied. As regards chemical survivability we have found that, in the presence of minerals, at temperatures up to 200°C the phenylethanol starts to decompose with formation of styrene, acetophenone, and chiral molecules such dibenzylethers and 2,4-diphenylbutanes. A tentative reaction mechanism is given.

ORGANIC MATTER IN HYDROTHERMAL SYSTEMS IN KAMCHATKA PENINSULA IN CONTEXT OF THE ORIGIN OF LIFE

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The geothermal environments on the early Earth are considered as the most suitable medium for the origin of life by many scientists. From this point of view, it is interesting to explore organic substance from hot springs inhabited by (hyper) thermophiles, and from lifeless water-steam mixture, where organic compounds could not appear as a result of destruction of modern microorganisms. To that end moderately volatile organic matter in water and water-steam mixture from the hot springs and boreholes in the 4 hydrothermal fields in Kamchatka was analyzed. Temperature of solutions in hot springs of Mutnovsky, Uzon and Karymsky fields is within the interval 70-99°C, pH 2.5-6.5. Water-steam mixture in the boreholes of Mutnovsky and Pauzhetsky fields is under the pressure 1.7-8.2
bars, temperature 108-175°C, pH 4-7; depth of the boreholes ranges from 600 to 2000 meters. To analyze the samples the GCMS-QP20105 Shimatsu was used. Phthalates were present in all samples, presumably as trace contaminants from plastic containers. The highest-temperature (175°C) sample from the borehole contained only polycyclic aromatic hydrocarbons (naphthalene, biphenyl, phenanthrene, fluorene, 1-methylnaphthaline). These organic compounds characterize the deep lifeless zone near the active Mutnovsky volcano (depth of the zone is 200-600 meters, temperature 175-250°C). Biphenyl and phenanthrene were absent in samples from the lower temperature boreholes (108-124°C). However, these samples contain some more aromatic hydrocarbons (benzenes, xylenes), as well as compounds of other homologous series: n-alkanes, aldehydes, ketones, alcohols. According to L. Mukhin (1979), glycine was detected in the lifeless water-steam mixture from Pauzhetsky field. Aromatic hydrocarbons, n-alkanes, alkenes and methyalkanes are present in hot springs of Mutnovsky, Uzon and Karymsky fields inhabited by the (hyper) thermophilic microorganisms. These data were integrated with the results obtained by other researchers in the framework of the inversion approach to the origin of life (Kompanichenko, 2008). According to the approach, initial prebiotic organic microsystems or aggregates were mainly composed of hydrocarbons, ethers, lipids or their precursors, amino acids (glycine) and some other compounds suitable for hydrothermal medium. Under the specific far-from-equilibrium conditions in hydrothermal medium the balance between the contribution of entropy and contribution of free energy (Scontr./Fcontr.) in the microsystem could become inverse that transformed it into the initial living unit (probiont). It is supposed that the synthesis of other biologically important molecules (sugars, ATP, nucleotides), which are not very typical for hydrothermal medium, started at the moment of the inversion Scontr./Fcontr. balance. The appeared at the moment negentropy motive power could facilitate the organized (non-spontaneous) synthesis of nucleic acid chains in combination with amino acid chains under higher temperature than in a chemical system in Vitro. This supposition is based on the comparison of maximal temperature limit of the nucleotide synthesis in Vitro (60°C, in the rare cases up to 90°C) and in Vivo (up to 110-120°C in the simplest hyperthermophilic Archaea).

References:

FLUCTUATING HYDROTHERMAL ENVIRONMENTS IN KAMCHATKA PENINSULA RELEVANT FOR THE ORIGIN OF LIFE

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The proposed inversion approach to the origin of life stated the necessity of fluctuating conditions in the medium for the origin of life (Kompanichenko, 2008). Hydrothermal environments suit this notion. Significant oscillations are peculiar to hydrothermal systems including their outcrops in ocean and terrestrial groundwater aquifers. The scale of the oscillations depends on the tectonic-magmatic and seismic activity of a geothermal region. Exploration of thermodynamic and physico-chemical fluctuations in natural hydrothermal fields can be helpful to base laboratory experiments on prebiotic chemistry under changeable conditions. To characterize a scale of the thermodynamic and physico-chemical fluctuations Mutnovsky and Pauzhetsky hydrothermal fields in Kamchatka geothermal region were explored. Temperature of water and water-steam mixture in the boreholes of Mutnovsky and Pauzhetsky fields ranges from less than 100 °C up to 240 °C. Data of monitoring of pressure and
temperature were mathematically processed. Period of long-range macrofluctuations of pressure and temperature in Mutnovsky field estimated on the 12 boreholes is between 2 and 4.5 months. The maximal amplitude of temperature on the wellheads is 53°C, the maximal amplitude of pressure is 34 bars. Coefficient of correlation between the temperature and pressure changes is between 0.88 and 0.99. The monitoring of short-range microoscillations of pressure in the particular boreholes in Mutnovsky and Pauzhetsky on the depth about 1000 meters led to the following result: period of the oscillations is within the interval 10-60 minutes, the average amplitudes of pressure are 0.2-0.7 bars. The general conclusion is that macro- and microoscillations of the thermodynamic parameters in hydrothermal systems are usual phenomenon. From time to time a scale of the fluctuations may significantly rise because of earthquakes and volcanic eruptions. Such changeable nonequilibrium medium seems suitable to be considered as potential geological cradle of life on the early Earth.

HOW STABLE WAS THE NITROGEN ATMOSPHERE OF EARLY EARTH?
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According to recent simulations of the Earth's nitrogen-rich thermosphere, the temperature of the upper atmosphere is not expected to rise above 7000 - 8000 K even under extreme solar EUV conditions anticipated for the early Earth. Rather, when the solar EUV flux exceeds some critical value, the escaping flow of the bulk upper thermosphere starts cooling it due to adiabatic expansion, which results in a decrease of the exobase temperature. Under these extreme solar conditions, the exobase may expand above the magnetopause and the magnetosphere will not be able to protect the upper atmosphere against strong non-thermal erosion by the solar wind. We show that a nitrogen-rich early terrestrial atmosphere with a present-day composition would have been removed within a few million years during the extreme EUV and solar wind conditions that are expected to have prevailed before the late heavy bombardment period about 3.8 Ga ago. Our results suggest that young Earth-like planets need either a CO2 amount which is about two orders of magnitude higher than the present-time level or a dense hydrogen-envelope which remained from the primordial atmosphere or the nebula so that an early nitrogen-rich terrestrial-type atmosphere could have been protected from complete destruction.

PLASMA AND CRATERS OF METEORITE IMPACT AND PREHISTORY OF LIFE
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A new idea of possible synthesis of the complex organic compounds in hypervelocity impact generated plasma torch were proposed and experimentally confirmed. Previously unknown and found experimentally new feature of impact generated plasma torch has allowed to developing the original concept of the prehistory of life. According to this concept the intensive synthesis of complex organic compounds arose during meteoritic bombardment in first 0.5 billion years at the stage of the planets formation. This the most powerful and destructive action in the Earth history could play the decisive role and prepare the conditions for origin of the life. In the interstellar gas-dust clouds the synthesis of simple organic compounds may be explained by identical process occurring in the plasma torch of hypervelocity collisions between the dust particles. It is assumed that the processes occurring in the
highly unbalanced hot plasma simultaneously with the synthesis of simple and complicated organic compounds were capable to ensure their ordering and assembling. Bona fide experimental evidence presented below indicates that the physical fields generated in the plasma environment in the process of the formation and expansion of the torch meet the main requirements toward “true” local chiral fields. These fields were very likely to be capable to trigger the initial, weak breaking of enantiomer’s symmetry and determine the “sign” of the asymmetry of the bioorganic world. These fields could have worked as “trapping” fields influencing spontaneous processes occurring in highly overheated and non equilibrium plasma in the state that is far from the thermodynamical branch of equilibrium and may have contributed to the formation of an environment needed for the synthesis of homochiralic molecular structures, which, in turn, were needed for the emergence of the primary forms of living matter. It has been shown experimentally that the plasma-chemical processes in the torch have high catalytic properties and assure the rise of the chemical reactions rates by 10-100 millions times. In the process of the plasma fly-away this in turn can assure fast forming the simple and complicated organic compounds including highly forked polymers. One may assume that predominantly inorganic substances from meteorites were used for synthesis of complicated organic compounds on the early Earth. Laboratory experiment with modeling the hypervelocity impact plasma torch by the laser working in Q-switch regime has shown the possibility of synthesis of high-molecular, ~5000 a.m.u, organic compounds by impact of micrometeorite with effective diameter 100 mkm. The target was composed of only H, C, N and O in inorganic form. The obtained of mass-spectra evidence to the high velocity of chemical reactions due to plasma catalytic processes. Some signs of self-assembly and ordering were observed. This allows to concluding that the plasma torch with huge local density of energy and matter may be the optimal medium for synthesis of complex organic compounds needed for the origin of the primary form of living matter. Having the giant energy, the meteorite impact is capable to inject the new-created complicated organic compounds deep inside the space body surfaces, including subsurface water reservoirs, such as, for example, on Europe, Enchilada and Titan. In this case the meteorite impact has no natural alternative in creation the initial conditions for origin of extraterrestrial life. Such a possibility was confirmed by laboratory impact modeling experiment, in which the plasma torch was created under the water surface. The proposed concept is based on real physical processes occurring in the nature and on experimental results of study the problem in impact experiments and modeling its analogues in laboratory conditions. Thus, the realizability and survivability of this concept should be taken as well grounded due to the simplicity and clarity of physical processes.
was a proof of spontaneous life generation will be described together with its conclusions and the influence they had on the further developments of wine-making and generally food industry. More generally, the twentieth century consequences on astrobiology and environmental sciences of Pasteur postulate that “life can only originate from life will be exposed.

FORMATION OF BIOLOGICALLY IMPORTANT MOLECULES ON THE MINERALS
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During the formation of planet high concentration of mineral particles generated due to high volcanic activity might be accumulated in the upper atmosphere. These mineral particles experience the influence of solar ultraviolet radiation and could change their composition under the action of light. On the surface of mineral particles a variety of photochemical transformation may occur. Here, the possibility of generation of complex biologically significant molecules on the UV-irradiated mineral surfaces was studied. We have shown, that on the surface particles of the minerals (clays and volcanic ash) preliminary irradiated by UV light there took place the formation of biopolymers from the adsorbed monomer molecules while in the absence of UV irradiated it did not occur.

THE EARLY STAGES OF LIFE ON THE EARTH
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Problems of appearance, formation (coming into being), early evolution of organic world and environments on the Earth are among basic problems of modern science. Two main conditions are noted in the history of every organism group development, comprising Earth biosphere. They are – appearance and wide expansion (or wide distribution). These stages don’t clash and be apart one from another for hundreds millions of years. The first appearance of new organisms group corresponds to certain level of temperature and atmosphere oxygenation; wide expansion as, for example, in the case of Metazoa corresponds also to the salt composition stabilization and to the “world ocean” volume or to some other events of the same scale. It is presumed that water on the Earth surface was absent or that it was minute amount of water in the period between 4.6 and 4.0 GA. In a great deal water appeared about 4.0 GA just after the last intensive meteorite bombardment. In this period sedimentary rocks begin their formation in shallow water basins and life roughly flourishes on our planet surface. It is not inconceivable/improbable even appearance of green algae and may be even fungi, besides bacteria, including cyanobacteria. Land surface also was inhabited by microorganisms already in Archaean. The appearance of cyanobacteria → eucaryots → Metaphyta → Metazoa happened much more early than it was proposed. The idea about atmosphere oxygenation level could be given according the level of organism organization. Water volume in oceans commensurable with modern one was formed about 1.3 GA. Owing to this fact the great expansion of different organisms is noted. Medium surface temperatures in Archaean cannot differ from modern more than 15-25oC. The world of RNA, if it existed, could be only before 4.0 GA or may be even before Earth formation. The chances of life appearance just (exactly) on the Earth is very probable. The study was executed within the framework of the complex program of basic research of the Presidium of the Russian Academy of Sciences “Origin of Biosphere and Evolution of Geo-biological Systems” (subprogram II) and was supported by the Russian Foundation for Basic Research, projects 08-04-00484 and SS-4207.2008.5.
A MODEL OF EARLY ATMOSPHERES PRODUCED BY IMPACTS ON THE EARTH AND MARS

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Existence of atmospheres is an important factor for the origin of life on planets, however, the birth of planetary atmospheres is still an unresolved problem. The Earth and Mars could have dense CO2 rich atmospheres in their early histories. A model of atmospheric evolution controlled by impacts, neglecting outgassing of solid planets, has been suggested by Zahnle [1] and applied for atmospheres of Titan and Mars. Atmospheric erosion by impacts has been taken into account using an analytical “tangent plane approximation”. The model was employed for the assessment of early terrestrial and martian atmospheres in [2] where the efficiency of atmospheric cratering was calculated for vertical impacts. Recently, accurate values of coefficients of atmospheric erosion and projectile material retention have been obtained by 3D hydrodynamic simulations of impacts [3]. Scaling laws derived in [3] and the model of impact-controlled atmospheric evolution [1] are used in this work for modeling of the evolution of the Earth’s and Mar’s early atmospheres at a late stage of planetary accretion. The atmosphere is considered to consist of two gases CO2 and N2. It is assumed that all carbon of vaporized fraction of impactors and targets is converted to CO2. Data on carbon and nitrogen abundances in stony meteorites and comets have been taken from the literature. Modern mass and impact velocity distributions of asteroids were used as an approximation for the late veneer period. It was assumed that the masses of largest impactors diminish in the course of accretion as follows from a statistical approach. For the early Earth, after the end of accretion, the model produces CO2-rich atmosphere with a pressure 20–100 bars and a partial N2 pressure 0.1–0.6 bars. The lower values were obtained neglecting cometary delivery of volatiles. The final atmosphere on Mars is more sensitive to input parameters of the model (like impact velocities, percentage of carbonaceous chondrites and comets, a fraction of volatiles released in the atmosphere) because of lower impact velocities on Mars and a smaller fraction of material vaporized and ejected to the atmosphere. On the assumptions that all the martian impactors are stony, 10% of them are carbonaceous, and that, on the average, 10% of a projectile mass is vaporized and the mass of vaporized target material is twice that of projectile vapor, the final atmospheric pressure is about 0.2 bars. And this pressure turns out to be about 5-10 bars on the assumption that a major part of carbon carried by stony impactors eventually reaches the atmosphere as CO2 or if a late veneer of comets is added. Large carbonaceous impactors are found to deliver more atmospheric components than the atmospheric mass lost after their impacts, while smaller impacts are more erosive. For this reason several random impacts of large volatile-rich bodies at the end of a late veneer could substantially increase the ultimate mass of an early martian atmosphere.

References:

A PUTATIVE WAY TO BIOLOGICALLY IMPORTANT METABOLITES VIA CATALYTIC SYNTHESIS FROM FORMALDEHYDE AND AMMONIA

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The modern ‘RNA-world’ theory considering the primary life as a system of self-replicating informational and catalytic oligomers is the most widespread hypothesis of life’s origin. At the same time an alternative major theory antithetic to the genetic one exists. It is based on the primary
formation of important metabolites and appearance of the primordial autocatalytic cycles in geochemical environment. [1]. Encouraged by this theory, we suggest the putative emergence pathways to biologically relevant metabolites from simple precursors: formaldehyde (FA) and ammonia, which are known to be present in the interstellar space [2]. Based on our earlier [3, 4] and novel findings as well as on the literature data [5, 6] the scheme of conjugate autocatalytic processes including photochemical condensation of FA yielding C-C bond and catalytic formation of pyruvaldehyde, pyruvic acid, sugars, aminoacids catalyzed by minerals (aluminum silicate, phosphates) and by aminoacids themselves is proposed. Acetaldehyde, glyoxal, glycol- and glyceraldehydes (GA and GCA) were identified in this work as products of photolysis of FA aqueous solution with summary selectivity up to 20% and can serve as substrates for the synthesis of more complex organic compounds. In turn, alanine and pyruvic acid are formed during UV-irradiation of aqueous solution of acetaldehyde and ammonium nitrate [6]. Pyruvaldehyde formation from GCA and FA in presence of amino acids was observed by A. Weber [5]. Finally, we revealed the catalytic activity of zeolite HZSM-5-17 in acid form towards formation of pyruvaldehyde from GA and GCA in presence of alanine with the yield about 5%. This observation indicates that the assumption of catalytic activity of silica-alumina towards the synthesis of metabolites could be realistic. Prebiotic synthesis of sugars from lower monosacharides, as we showed earlier, could take place over phosphate catalysts [4]. The substrates and the products of the proposed scheme were detected in the interstellar space and meteorites [7, 8], evidencing of the possibility of its realization either on the Protoearth or in the protoplanet disk supplying the initial compounds for contemporary metabolism.

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References:

THE TEMPLATED PHOTOCHEMICAL PROCESSES IN PREBIOTIC EVOLUTION
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The photochemical processes, which reactions proceed along the excited-state pathway are considered as an important source of organic molecules in prebiotic chemistry and often lead to products virtually inaccessible by thermal reactions. One of present trends in photochemical experiments targeted to reproduce prebiotic processes is the investigation of templated processes instead of reactions proceeding in homogenous solution or a gas mixture. By template we mean a host molecule containing a sensitizer which reversibly binds a substrate molecule and does not become part of the product. Besides increasing local concentration of reagent molecules and directing their spatial arrangement, the template should act as a catalyst thus favoring the effectiveness of reaction. Prebiotic evolution of organic molecules was closely linked with an evolution of templates starting with mineral surfaces of silicates and aluminosilicates through abiogenic organic matrices such as melanoidins and pigmented amino acid polymers (proteinoids) to highly directed and efficient molecules of genetically ordered biopolymers. To illustrate the role of templates in abiogenic synthesis of biologically important compounds we will discuss the role which mineral (titanium dioxide, silica gel) or organic (melanoidin) matrices played in the formation of organic acids, amino acids and peptides under the action of UV radiation. In the context of evolution of
prebiotic templated photoprocesses two models of abiogenic ATP synthesis were designed and studied in our laboratory. One of these models sensitized photophosphorylation of ADP molecules which had been adsorbed on montmorillonite particles and then subjected to UV-irradiation. The other system was based on the photochemical activity of izoalloxazine (flavin) and pteridine pigments conjugated to proteinoid structures formed after a thermolysis of anhydrous amino acid mixtures. The results of spectroscopic study to elucidate chemical structure of photochemically active abiogenic pigments and the factors influencing the effectiveness of templated systems of ATP synthesis will be discussed.

POSSIBLE “NIDI” OF ORIGIN AND SURVIVAL OF MICROORGANISMS IN THE PROTOPLANETARY CIRCUMSOLAR DISK
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The current standard model of the Solar system origin is submitted. Here we consider the physical and chemical conditions in the first 100 million years of formation of the Solar planetary system in the near zone (terrestrial planets) and distant zones – the zone of giant planets and their satellites, ice nuclei of Jupiter-family comets and bodies of the Kuiper belt, scattered disk and Oort cloud. The conditions on the surfaces of primary Venus, Earth and Mars, forming in the so-called zone of possible life are considered. The calculations for trans-Neptunian bodies showed the possibility of formation of subglacial water reservoirs (the oceans) with organics. Estimated time of their existence is about a billion years. The results of the calculation are presented in diagram form “P (pressure) – T (temperature) – concentration of water – concentration of organics”, which give an indication of the possibility of the origin of life in the near and far fields of the young solar system.

References:

GROWTH, UVC RESISTANCE AND DIFFERENTIATION OF BACILLUS SUBTILIS STRAINS ISOLATED FROM AN EVOLUTION EXPERIMENT
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Solar UV radiation drives essentially all life processes at the Earth’s surface, but paradoxically it is also a major source of lethal damage to cellular components. In a precursory study for the space experiment ADAPT (Molecular adaptation strategies of microorganisms to different space and planetary UV climate conditions) we studied the adaptiveness of Bacillus subtilis to periodically applied polychromatic UV irradiation. The utilized spectrum (200 - 400 nm) models the suggested UV radiation environment on the early Earth, which lacked a significant ozone layer. As we reported previously, stationary phase cells of UV evolved populations were about 3-fold more resistant compared to the ancestral and non-UV evolved populations. However, spores of all populations respond similar to UV irradiation and exhibit ancestor-like survival characteristics. It is known that variations in the UV sensitivity of cells exist during growth in batch cultures. Therefore we performed UV characterization experiments at several defined stages of growth and differentiation.
Additionally, we analyzed other phenotypic traits that have changed, were evolved or were lost in the course of the experimental evolution, e.g. decreased sporulation efficiency. Although the molecular mechanisms behind most of the observed changes are not found yet, it is of interest if the loss of one trait could have facilitated the adaptive evolution of another or if they are correlated in another way.

**Theme 2: Life in extreme environment**

**NEW THERMOPHILIC PROKARYOTES WITH UNUSUAL METABOLIC FEATURES FROM TERRESTRIAL AND DEEP-SEA HYDROTHERMAL ENVIRONMENTS**

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Phylogenetic and metabolic diversity of thermophilic prokaroyotes is intensively studied during past three decades, and many new, metabolically diverse microorganisms were isolated; some of them represented deep phylogenetic lineages both in Archaea and Bacteria domains. Many of these organisms are thermophilic and hyperthermophilic lithoautotrophs growing at the expense of anaerobic oxidation of inorganic substrates of volcanic origin. Still, direct phylogenetic analysis of thermophilic microbial communities demonstrate the presence of many new organisms not obtained in laboratory cultures. The goal of our work was the isolation of new thermophilic microorganisms using inorganic energy sources in the process of anaerobic growth.

From the hot springs of Kamchatka Peninsula, Russia, strains of new hyperthermophilic bacterium growing optimally at 80°C were isolated and described as a novel genus and species *Caldimicrobium rimae*. This organism belongs to *Thermodesulfbacteria* phylum and can grow lithoautotrophically with molecular hydrogen reducing elemental sulfur or thiosulfate. Strains of *C. rimae* are also capable to oxidize volatile fatty acids and alcohols – the fermentation products of organotrophic hyperthermophilic Archaea and Bacteria.

Another new isolate, also representing a new genus in phylum *Thermoproteobacteria*, was obtained from the deep-sea hydrothermal samples of Lau Basin, Pacific Ocean. New organism is an obligate lithoatrotroph growing at 92°C on the mineral medium by dismutation of sulfur compounds – elemental sulfur or thiosulfate, during one molecule is oxidized to sulfate and another reduced to sulfide. The growth is obligately dependent on the presence of ferric oxide in the medium, which binds sulfide formed in the course of growth, maintaining its low concentration in the medium.

CO is a usual component of volcanic gases, both in terrestrial and submarine hot springs. The ability to grow anaerobically at 100% CO in the gas phase producing molecular hydrogen and CO₂ was found to be widely spread among thermophilic prokaryotes – Bacteria of phylum *Firmicutes* and members of archaeal genus *Thermococcales*. However, if the concentration of CO in gas phase was 5 to 45%, the range of microorganisms capable of hydrogenogenic CO-trophy became much wider. Among new organisms capable of this type of metabolism are hyperthermophilic bacteria of *Dyctioglomy* phylum and hyperthermophilic crenarchaeote 'Thermofilum lithoautotrophicus'.

These and some other new thermophilic lithoautotrophic microorganisms able to use energy substrates, electron acceptors and carbon source of geothermal origin can act at the base of microbial food web not dependent neither of solar energy, nor of the modern biosphere.

**MICROBIAL DIVERSITY IN SALT LAKES OF NOVOSIBIRSK REGION. MOLECULAR IDENTIFICATION AND BIOTYPING**


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The microbial communities developing in inland water bodies present considerable interest from point of biosphere evolution and some researchers believe that they are analogues of ancient Earths communities (Zavarzin, 2007). These ecosystems are unique both natural biotopes and sources of biotechnological perspective strains (Oren, 2002). The microbial communities of salt lakes of the Novosibirsk and Altai regions were studied in a complex work simultaneously with the study of geochemical condition of microorganism’s habitat. The structure of microbial communities was studied in water and sediments samples by technique of fluorescence in situ hybridization (FISH). Phylogenetic position of isolated strains was detected by molecular biological methods (16S rRNA gene sequences). Individual protein profiles (spectrums) were obtained by a method of time-of-flight MALDI TOF mass spectrometry and treated in the specialized program BioTyper. The water of investigated lakes subdivided into 2 types – chloride and chloride-sulfate. Most lakes of chloride-sulfate type has salinity level from 200 to 250 g/l. Waters of few lakes has lower salinity (7 – 50 g/l). Values of pH vary from 7 to 9. At the microscopic analysis of structure of planktonic microbial communities have been found that all three domains of life: Bacteria, Archaea and Eucarya are widely presented in the investigation lakes. In three lakes development of benthic microbial communities of halophilic cyanobacteria and eukaryotic algae were marked. Archaea pigmented lakes waters in pink-violet color in the case of their mass development. In natural samples at salinity over 230 g/l archaea prevail above other forms: bacteria, cyanobacteria and algae. At salinity more than 250 g/l number of archaea began to decrease. Laboratory experiments confirmed that the optimum of development of archaeal strains is 250 g/l NaCl. Most bacterial cultures grow at low values of NaCl (less than 200 g/l) and they did not grow at salt concentration more than 250 g/l. However they grow observed at absolute absence of NaCl in a medium. From the samples of water and bottom sediments more than 200 bacterial and archael strains belongings to the different ecological and physiological groups were isolated. Phylogenetic position some of them were established. These organisms were halophilic bacteria and archaea belong to genera Haloarcula, Halorubrum, Halomonas, Salicola at alias. For the some isolates individual protein profiles were obtain and analyzed. It is very important to note that phylogenetic tree which was built on the basis of mass spectrometry analysis was a similar with tree which was built by a method of the minimum-evolution (ME). Research of structure of microbial communities of Novosibirsk and Altai regions salt lakes have shown that salinity and pH were the key factors in determining the composition of the microbial communities. Communities which were investigated represent an interesting example of a diverse communities of halophilic bacteria, archaea, cyanobacteria and algae well adopted to a broad salinity range. This work was supported by the RFBR 08-04-01798 and 08-05-00968; IP 10 and 73.

DIURNAL HABITABILITY OF FROZEN WORLDS

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In this work we discuss effects allowing local habitability of some extraterrestrial planets despite of low average surface temperature. We analyze the problem of diurnal and seasonal changes of temperature and biological productivity at different locations on a hypothetical Earth-like planet. Under some circumstances the temperature may locally rise well above the average value, allowing periods of enhanced biological activity. In this way, bioproductivity can become periodically possible on a planet that has average temperature clearly below 0°C. Such thermal conditions are encountered on Mars, generally considered as inhabitable. In reality, an appropriate temperature is not sufficient for habitability. The presence of liquid water at the considered location is also necessary. We discuss
how temperature oscillations affect habitability in the framework of a conceptual model. Our results may be important for Earth-like planets situated near the outer edge of the HZ. In this case our previously applied definition of habitability [1] should be generalized: An Earth-like planet may be diurnal habitable if at least some special regions of the planetary surface have non-zero biological productivity during certain time intervals of the year. This effect depends strongly on the obliquity. It should be noted, that the effect of diurnal habitability discussed in our paper can allow only the preservation of primitive organisms as observed in terrestrial polar cold deserts [2].

References:

BIOGEOCHEMICAL INVESTIGATIONS AND MICROBIAL BIODIVERSITY IN LAKE UNTERSEE AND OTHER PERENNIAL ICE-COVERED ANTARCTIC LAKES
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The freshwater, low-mineralized, and hypersaline perennially ice-covered lakes of the McMurdo Dry Valleys (1995–1996), as well as the freshwater, ultra-oligotrophic, hyper-alkaline Lake Untersee (Novolazarevskaya Station, 2008), were investigated. The data obtained in 2008 by the Tawani Foundation Antarctic International Expedition indicate plenty of microorganisms for thorough investigation for biogeochemistry and biodiversity in the Lake Untersee. In the water column of these lakes, we determined the rates of photo- and chemosynthesis, sulfate reduction, methane oxidation and methane production, and 3H thymidine incorporation. The concentrations of oxygen, methane, organic carbon, nitrogen compounds, ATP, and chlorophyll were measured as well. The highest rates of the biogeochemical processes of photo- and chemosynthesis and methane oxidation were detected in the water column of the chemocline; the highest rates of sulfate reduction and methane production were observed in anaerobic horizons and bottom sediments. In Lake Untersee, the proteobacterial phylotypes related to heterotrophic and photosynthetic groups of organisms belonging to the genera Hyphomonas, Phenyllobacterium, Polaromonas, Roseococcus, and Sphingopyxis were found in the 16S rRNA clone libraries obtained from 60-m, 80-m, and 98-m horizons. For Lake Fryxell, a series of mathematical models simulating oxygenic and anoxygenic photosynthesis, sulfate reduction, methane production and methane oxidation, as well as models for methane, sulfate, oxygen, and organic carbon concentrations, were developed. For Lake Untersee, mathematical models describing the same processes and the concentrations of the aforementioned compounds are to be developed using the data obtained in the course of the expeditions conducted in 2008 and 2011.

CRYOBIOSPHERE: TERRESTRIAL ANALOGUES AND MODELS OF MARTIAN HABITATS AND INHABITANTS
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Terrestrial Cryosphere, inhabited by viable microorganisms, represents a range of possible extraterrestrial cryogenic ecosystems on the Earth-like planets such as Mars. In its balanced environment cells survive significantly longer than in other habitats. If life existed during the early stages of Martian development, then remnants of primitive forms may be found within frozen material that protects them against unfavorable conditions. The paper considers: (a) Icy World - ice sheets as the Earths most representative analogues of ice caps on Martian poles, (b) the suggested age of Antarctic permafrost as an analogue somewhat closer to that of Mars than Arctic permafrost, (c) free water on Mars existed as cryopegs - brines formed when Mars became cold, (d) volcano-ice interactions in permafrost areas as one way to have liquid water and life on Mars, (e) young volcano permafrost on Mars which age somewhat closer to terrestrial one and (f) soil cover as a distant model of the Martian “active layer”.

NEW VIRUSES IN HYDROTHERMAL DEEP-SEA ECOSYSTEMS

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Deep-sea hydrothermal vents represent one of the most extreme environments on Earth. These ecosystems are characterized by very high pressure, the lack of solar energy and the prevalence of chemosynthesis. Our laboratory is specialized in the study of viruses isolated from deep-sea hydrothermal heterotrophic sulfur-metabolizers, is indeed one of the predominant groups of the hyperthermophilic microbial communities from deep-sea vents. We performed a systematic search on samples collected in various geographically hydrothermal sites located on the East pacific Rise, Mid-Atlantic Ridge and Central Indian Ridge. This study, on enrichment cultures, revealed an unexpected viral diversity. Among the many morphotypes observed, the lemon-shaped type prevailed (Geslin et al., 2003), In parallel to the diversity, only one virus-like particle isolated from a marine hyperthermophilic Euryarchaeota was described: PAV1 isolated from Pyrococcus abyssi strain GE23. (Geslin et al., 2003) It displays unique features at the nucleic and proteinic level (Geslin et al., 2007) Recent studies were led to isolate 3 new viruses: TV1 for Thermococcus Virus 1, TV2 (Thermococcus Virus 2) and PAV2 (Pyrococcus abyssi Virus 2). These viruses are lemon-shaped (140 nm, 120 nm and 160 nm respectively), resemble PAV1 and all possess dsDNA genomes. Viruses, the most abundant entities on the planet, affect the three domains of life and are detected in the most “extreme” environments. They have probably influenced the Origin of Life (Raoult et al., 2008; Rohwer et al., 2009; Prangishvili et al., 2009).

MODIFICATIONS OF THE STANDART METHODS OF BACTERIAL REACTIVATION FROM PERMAFROST SEDIMENTS

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The most complicated problem in searching of microbial life in extreme environments (both terrestrial and extraterrestrial) is to reinstate the growth of the cells. It has been known that only a very small fraction of the microorganisms present in permafrost sediments can be enumerated by standard plating methods on nutrient media. The discrepancy between the total number of cells and the number of culturable cells can be explained by entering of bacteria to resting forms, or non-culturable state and formation of “dwarf” cells, the latter in most cases incapable to produce colonies on standard media. All variants of physiological state need various procedures of cell reactivation
Physiological diversity of permafrost bacteria in situ is revealed by different levels of cell reactivation on nutrient media and possible depends on extended time of freezing in sediments. The aim of present study was to improve resuscitation procedures of bacteria from permafrost sediments for more complete isolation of viable bacteria. Methods of reactivation of dormant cells were studied both in native bacterial populations and resting bacterial forms obtained in model experiments, and included washing off the microbial fraction from growth inhibitors and other factors that sustain cells in anabiosis. Besides, in accordance with recent concept that physiological state of the cells in microbial populations is likely to be controlled by intercellular communications provided with signal molecules, the standard methods of isolation were modified by adding the following regulatory factors: autolyzed yeast as biological active substance and nutrient; alkloxybenzol (C₇-AOB) as microbial autoregulator of growth and antioxidant; wheat corcule agglutinin and plant hormone -β-indole-3-acetic acid as molecular signals for growth. Suggesting methods provide the possibility to increase resuscitation of bacterial growth in extreme Earth habitats and may be useful for search life on extraterrestrial bodies.

**METHANOGENS IN PERMAFROST**

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Martian permafrost might hypothetically record genetic signatures of preexisting life, which has long since vanished. Best Earth’s analogue of Mars is permafrost, which studying gives an understanding of our possibilities to find life on Mars. Among the various microorganisms observed in the permafrost, methanogenic archaea is one of very important group of anaerobic lithotrophs. The main goal of this work was the investigation of methanogens in Arctic permafrost. We also tried to get answer how methanogens have been preserved during geological time and maintain viability. Samples from typical Arctic permafrost horizons of different age (modern, 10 and 20 to 50 Kyr and 0.6 to 3 Myr) and genesis were studied. Culture and culture-independent methods were employed to characterize the culturable and unculturable microbial populations. Three cultures of methanogenic archaea were isolated from Holocene and late Pliocene horizons. New species Methanobacterium veterum sp. nov and strain Methanosarcina mazei JL01 were isolated and described. tDNA were extracted and amplified with archaea-specific primers. Positive results were observed with tDNA from modern soil to 50 Kyr old permafrost samples. It was shown domination of order Methanosarcinales in majority of analyzed permafrost samples.

**PHENOMENON OF THE BLACK SEA SMOKER**

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In 2001 from a bottom of Black sea the fragment of the smoker has been lifted from depth 1600 m. The raising has been carried out by a drag, the sizes of fragment: 1.0 x 0.5 m. The external part chimney has been covered by organic gel in the thickness of 10 mm. The basic part of the lifted smoker is exhibited at Central Museum of Natural History in Kiev. The object owns the astrobiological aspects due to extreme living conditions of bacterial floor-mats which cover outside wall. It has been carried out radio carbon dating. Element, isotopic, mineral and microbiological
THE BIODIVERSITY OF AQUIFERS IN THE SUBSURFACE – A SPECIAL TREASURE ON PLANET EARTH

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Water is a most fundamental issue in astrobiology with regard to understanding the principles of the biosphere on our planet, as well as with regard to the search of past and/or present biospheres in other parts of universe. What lessons can be learnt from the aquifers below our feet that penetrate nearly our whole subsurface on our planet? Principally, the aquifers harbour at least two treasures for various life forms on our planet: they constitute invaluable groundwater resources for higher life forms; and they serve as a dynamic, mobile subsurface reservoir for a yet largely unexplored constantly evolving pool of various microorganisms. However, given the large amount of diverse aquifers on our planet and the difficulties to approach these with adequate methodology to assess microbial in situ activities without introducing misleading biases, few systematic comparative global studies are available. We have embarked on a holistic approach aiming at mapping the biodiversity in a selected amount of pristine and contaminated aquifers of different ages (down to >7,000 years aquifers) and geochemistries in Germany, Sweden and USA, with a focus on areas with dechlorinating and metal/radionuclide transforming species. For this, different methodologies were employed such as different statistical parameters and systematic evaluations of biogeography of different key groups compared to other ecosystems on our planet. Based on this information, emerging insights into the hidden diversity of our aquifers were made, resulting in the identification of several key groups of microorganisms such as novel candidate phyla and groups within deep branching phyla that enable us to frame fundamental questions on the origin of the biosphere in the subsurface and its constant impact on all surface-associated ecosystems on our planet.

ASTROBIOLOGICAL LESSONS FROM THE ANCIENT IRON-OXIDIZING GENUS GALLIONELLA

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The biology of iron transforming microorganisms is of fundamental importance for our understanding of past and present biogeochemical processes on Earth, as iron is the most abundant element. This abundance promoted a fascinating evolution of a diverse group of prokaryotes metabolizing different iron compounds in various ecosystems, encompassing chemolithotrophic and heterotrophic species, as well as different types of extremophiles from both prokaryotic domains. Iron metabolizing prokaryotes like the iron-oxidizing Gallionella thus constitute interesting models for our emerging comprehension of past and present prokaryotic evolution in iron-based environments and its current role in our biosphere as well as in astrobiology in general. Not only does Gallionella species belong to one of the few current species whose ancestors can be traced back to the eldest prokaryotic fossil records on our planet [Hofmann et al., 2008, Astrobiology, 8, 87], they still play a fundamental role in many ecosystems, e.g. in the vast unknown subsurface. Although the genus Gallionella was already described in 1836, our knowledge about its biodiversity is scarce. So far, only one strain, Gallionella capsiferriformansT, has been deposited in a type culture collection and just recently the draft for the genome sequence of Gallionella ferruginea subsp. capsiferriformans ES-2 was released. However, a recent inventory of hundreds of 16S rRNA gene sequences affiliated to Gallionella suggests that an unexplored diversity exists. Here, we describe our efforts to explore the global biodiversity of Gallionella in comparison to other iron-oxidizing genera, and to compare the genome of Gallionella strain ES-2 with other so far sequenced iron-oxidizing genera, and how this can reveal features that may lead to the development of analogous hypotheses on other iron-based biospheres in other parts of the universe.

GENETIC CHARACTERISTICS OF ANCIENT PERMAFROST BACTERIA AS COMPARED WITH MODERN-DAY SOIL AND WATER BACTERIA

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Preservation of viable microbial communities in permafrost sediments of different age testify, that Earth ancient biotopes with stable long-term subzero temperatures are characterized by balanced specific mechanisms, that provide cell structure stability and a high vitality of living system as a whole. Although much has been reported about bacterial resistance in frozen habitat, we are far from a complete picture about genetic resistance of bacteria under native conservation in permafrost. Bacterial isolates from permafrost give us a unique opportunity for direct molecular comparisons between ancient and modern-day bacteria which are experience anthropogenic stress. Antibiotic and mercury tolerance were analyzed for bacteria isolated both from Arctic and Antarctic permafrost and bacterial isolates from modern-day soil and water samples. The content of resistant strains in native samples, the taxonomic diversity of mercury and multidrug resistant ancient and modern-day bacteria, the genetic structure of resistance determinants and their association with different mobile elements were studied. Comparative analysis of the spectrum of antibiotic and mercury resistance, genetic characteristics of both ancient and modern-day bacteria revealed similarity in the level and resistant spectrum among the strains within one systematic group of bacteria. Besides, the strains from both collections revealed similar mobile elements (plasmids and transposons), where resistance genes are located. Thus, our results demonstrate that resistance genes were spread in natural bacterial populations long before the intensive introduction of antibiotics and mercury into the environment as anthropogenic stress. In addition, resulting genetic resistance in ancient bacteria point at existence of
mechanisms, that may prevent the accumulation of genetic damages in cells under long term freezing in Earth permafrost sediments. It seems possible to extrapolate these data on probable conservation of living forms on extraterrestrial cryogenic bodies.

BIOTECHNOLOGICAL POTENTIAL OF SIBERIAN PERMAFROST

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Extremophilic microorganisms represent the potential source of proteins with different functions and original characteristics not inherent to their analogues from other sources. The study of proteins from cold-adapted bacteria may provide a clue to the properties of extraterrestrial life on Mars and other cold celestial bodies. Specifically, bacteriorhodopsin-based light energy utilization mechanisms are widespread in different extremophiles and therefore constitute the promising target in astrobiology studies. Siberian permafrost contains the unique microbial community adapted to extreme conditions including long-term freezing, cumulative radiation level and high water osmolarity. It is inhabited by heterotrophic oligotrophic bacteria, Archaea, cyanobacteria, green algae, yeasts, mycelial fungi. Recently genomes of Exiguobacterium sibiricum, Psychrobacter cryohalolentis and Psychrobacter arcticus were elucidated by DOE Genome Institute. The presence of potential bacteriorhodopsin gene was predicted in the genome of E. sibiricum. This protein was expressed heterologously in E. coli membrane with yield of 10-15 mg/L culture. The primary investigation of its functional characteristics revealed interesting features which can be useful for the construction of biooptoelectronic devices. Several genes coding for potential enzymes (lipases, laccases etc.) with homology to known biocatalysts were found in the genomes of Psychrobacter and Exiguobacterium. Their expression in E. coli and functional characterization will contribute to estimation of biotechnological potential of Siberian permafrost as the extraterrestrial model. The work is supported by Federal Targeted Program on Scientific and Pedagogical-Scientific Staff of Innovative Russia.

DEVELOPMENT AND APPLICATION OF A REAL-TIME PCR METHOD FOR CHARACTERIZATION OF PERMAFROST ANAEROBIC MICROBIAL COMMUNITIES

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Permafrost is characterized by constant subzero temperatures and low (from 40 to -256 mV) redox potential. Such conditions have created possibilities for existence specialized anaerobic microbial communities. Difficulty of interpretation of the permafrost anaerobic microbial biodiversity data received by molecular ecological methods is defined by the low contribution of anaerobic microorganisms in the maintenance of total DNA from frozen grounds. Real-time PCR (QRT PCR) is a fast and highly sensitive method for the quantification of microorganisms from different samples.
Although the method is actively used in microbial ecology, its application for the analysis of methanogenic communities is limited. The purpose of this research was to evaluate the quantity of methanogens in the anaerobic microbial communities from permafrost with SYBR Green and TagMan PCR in real-time. The estimation of specificity picked up primers to anaerobic microorganisms has been spent for optimization the quantification of the microorganisms participating in the methane formation in permanently cold ecosystems with use of QRT-PCR method. Amplification of total DNA from five Arctic permafrost samples of late Pleistocene sediments in real time has shown that the total number of bacteria in investigated samples was 106-107 cells/g. Sulfate reducing bacteria have not been found, and methanogenic archaea (1.3x105 cells/g) there were only at one investigated sample from the depth of 17.5-17.6 m. More detailed research of DNA from this sample with the using group specific primer systems has shown the presence of two dominating groups of methanogens: Methanobacteriales and Methanosarcinaceae. The first results showed that application QRT-PCR allows to estimate not only the quantity of microorganisms of certain function in natural microbial community, but also the efficiency of selective conditions in enrichment cultures.

GAINING INSIGHT INTO SURVIVAL STRATEGIES OF BACTERIA IN FROZEN TERRESTRIAL ENVIRONMENTS

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Ancient Arctic and Antarctic permafrost is the regarded model for solving some important issues, relevant to astrobiology, concerning boundaries and mechanisms of microbial persistence in cold environments and trials of approaches to detect alive cells or biomarkers (if present) in frozen extraterrestrial habitats. Basing on analysis of our experimental data, it is possible to consider long-term exposure to subzero temperatures as a factor, with which only resistant microbial cells might cope in terrestrial permafrost. The existence of diverse microorganisms in ancient permafrost is evidently not limited to innate psychrophily of some species, since mesophilic prokaryotes are commonly isolated from these habitats. Abundant bacteria, found in the Earth permafrost, have adopted perhaps common mechanisms that ensured stability of cells, subcellular structures and genetic material for thousands and million years. In this communication we discuss properties of permafrost bacteria (ex situ and in situ) to be useful as targets or prototypes in further searches for life signs on extraterrestrial cryogenic bodies. A special attention is paid to the following facts.

1. Bacteria in permafrost samples, like the cultured cells of strains isolated from these cold habitats, preserve the structural integrity and viability after exposure to various external stresses.
2. Cells in permafrost are similar in the structure and physiological properties to dormant cystlike cells in laboratory cultures of non-spore-forming bacteria, including the isolated strains. Cystlike cells are responsible for survival of non-spore-formers in Earth permafrost and may be considered as targets in exobiological explorations.
3. Resistance of permafrost microbial communities to heating, osmotic shock, oxidative stress, and radiation can be due to inherent stress resistance of cystlike cells augmented by additional protection from their environment (organic and mineral particles, EPS).
In the experiment PUR 16 samples were exposed to the irradiation on the top of the compartment under appropriate neutral density (MgF2) filters according to the results of the simulation experiments. Under the exposed samples the dark samples were accommodated, these were protected from the solar radiation but the temperature oscillation, vacuum and particle radiation can influence them. Each sample holder was vacuum tightly closed containing inert Argon gas and sealed. A third series of samples is stored in Cologne in the Planetary and Space Simulation Facility aiming to perform the Mission Ground Reference Experiment of EXPOSE-R at DLR, while the fourth one is in the Laboratory for Biophysics, Budapest, Hungary. In addition to the flying dark control samples, two types of Ground Based Control Experiments are under performing: 1. In Cologne, in the Planetary and Space Simulation Facility at DLR 2. In Budapest, in the Laboratory of Research Group for Biophysics (RGB) Ad 1. In the Planetary and Space Simulation Facility the Mission Ground Reference Experiment of EXPOSE-R at DLR are performed, the samples are exposed according to the data obtainable from the ISS. However, the flow of data was several times interrupted because of technical reasons. The temperature data March—June, 2009 was available from June and the copied data were fed into the Ground Reference System of DLR. After installation of the telemetry system (December, 2009) the regulation of the Ground Reference System in Cologne takes place by these data – with exception the functional defect of telemetry. Ad 2. The temperature data in EXPOSE-R are incomplete because of the breakdown and exchange of the station PLC. The effect of the temperature variation on uracil thin layer was tested in RGB between + 20 and -20 degree of Celsius. The effect was measured both by UV spectroscopy and IR (FTIR) spectroscopy. After UV irradiation at the characteristic wavelengths of the uracil spectra a change was obtained indicating a change of the crystalline structure of uracil. The aforementioned spectral changes recovered in a short incubation period. These results we have to take into account in the on-line UV dosimetry planned by our group. The irradiation data were estimated according to the data of the returned EXPOSE-E facility.

EXAMINATION IN THE SIGNATURES OF BIOLOGICAL AEROSOLS IN THE EARTH ATMOSPHERE USING FTIR TECHNIQUE

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The passive remote spectrometric methods are important in examinations the composition of the atmospheres. The spectral technology can be useful in detection of the trace aerosols like biological substances (if present) in the environments of the planets. We discuss here possibility of detection and identifications of biological aerosols with a passive InfraRed spectrometer in an open-air

Numerically simulated, based on radiative transfer theory, spectroscopic observations of the Earth atmosphere were shown. Using measured values of the refractive index for bacterial spores, and assuming their size distributions, the single scattering albedo, optical properties such as extinction/absorption coefficients, transmittance was computed for bioaerosol clouds at a resolution of 2cm$^{-1}$. Then the calculation of radiance for a cloud immersed in the atmosphere using the radiative transfer code was done. Results of this simulation were compared with several measurements in laboratory testing tube and using our newly constructed FTIR spectrometer. Our theoretical and experimental studies indicate that, for a passive remote sensing measurement, it is difficult but possible that bioaerosol clouds can be identified from the spectral signatures.

**ORGANISM/ORGANICS EXPOSURE TO ORBITAL STRESSES (O/OREOS) NANOSATELLITE**

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The O/OREOS Sat mission is scheduled to launch on September 1, 2010 from Kodiak, Alaska on a Minotaur IV rocket. O/OREOS Sat carries two experimental payloads exposed to the space environment: (i) dormant biological specimens and (ii) four types of reaction cells containing organic molecules. O/OREOS Sat investigates the stability of organic material in space and the survival, adaptation, and biological evolution of life. Halorubrum chaoviatoris and Bacillus Subtilis spores (each as wild-type and mutant) will be launched in the dry state. During space flight, nutrient media will be added to sub-groups of organism-containing microwells at three timepoints over the 6-month course of the mission. Biological samples are monitored by time-resolved optical density and colorimetry of a metabolic indicator dye to quantitatively measure the effects upon biological organism survival, growth, and metabolism resulting from the combined exposure to ionizing space radiation and microgravity. In addition, O/OREOS will expose four classes of organic molecules to the space environment: amino acid, quinone, polycyclic aromatic hydrocarbon, and metallo-porphyrin. One of each specimen will be maintained in self-contained sample environments representing interplanetary space, airless bodies, and Mars environment. The main objectives are to use changes in UV and visible absorption spectra to quantitatively measure the effects upon organic specimens of the combined exposure to space radiation and UV and visible light. We report on how O/OREOS Sat will address the stability of organic and biological material in a variety of astrobiologically relevant space environments with in situ technology on small satellites.

**ASTROBIOLOGY ON THE INTERNATIONAL SPACE STATION**

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The environment of the International Space Station ISS provides a complex spectrum of physical parameters that are not experienced on Earth and that are of high interest to astrobiology. These parameters have been used in experiments on board of the European EXPOSE facilities attached to
the outside of the ISS (Rabbow et al. 2009). The chemical set of experiments has been designed to reach a better understanding of the role of interstellar, cometary and planetary chemistry in the origin of life (Brack et al. 1999, Cottin et al. 2008). From studies on the chemical evolution, survival, destruction and modification of complex organics, e.g., PAHs, fullerenes and complex aromatic networks in outer space experimental clues are obtained on the photochemistry of these compounds in the interstellar and interplanetary medium. Finally the chemical experiments contribute to the understanding of the chemical processes on Saturn’s moon Titan and possible analogies to the prebiotic chemistry on the early Earth. The biology experiments use the full extraterrestrial spectrum of solar UV radiation and suitable cut-off filters to study both, the role of the ozone layer in protecting our biosphere and the likelihood of resistant terrestrial microorganisms and microbial communities to survive in outer space. The latter studies provide experimental data to the hypothesis of lithopanspermia (Nicholson 2009), i.e. the interplanetary transfer of life via meteorites, and they provide basic data to planetary protection issues, i.e. the need to prevent contamination of target planets, e.g. Mars by terrestrial microorganisms. To get better insight into the habitability of Mars, samples are also exposed to simulated Martian conditions (UV-radiation climate, pressure, atmosphere), with and without a protective cover of simulated Martian regolith. The biological test samples selected are hardy representatives of bacteria, Archaea, lichens, fungi and plant seeds, i.e. of various branches of life, also in their natural communities. Most types have already demonstrated their resistance to outer space during short term missions, e.g. on board of the ESA BIOPAN facility (Horneck et al. 2001, Rettberg et al. 2004, Sancho et al. 2007, de la Torre et al. 2010). References:


CHEMOLITHOTROPHIC MICROORGANISMS AND MICROBIAL COMMUNITY FROM THE DESERT SOIL AS CANDIDATES FOR THE PHOBOS-GRUNT SPACE RETURN MISSION

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Three cultures of chemolithotrophic microorganisms and the desert soil have been selected for the biological experiment in the frame of the Phobos-Grunt space return mission. The goal of the experiment is to test the panspermia hypothesis, which posits that microorganisms have traveled between planets sheltered deep inside space rocks. The samples should be placed into a special container that will be attached to the spacecraft outside. In its flight, the container will serve as a simulated space rock, subjected to the same extreme conditions as a Martian meteoroid drifting to Earth. During the mission, the samples will be exposed to the factors of a deep space, beyond the protection of Earth’s magnetosphere for about three years. The cultures are two archaea Methanosarcina lacustris and Ferroplasma acidiphilum as well as a homoacetogenic bacterium Acetobacterium tundrae. Methanogenic archaea and homoacetogenic bacteria are autotrophic anaerobes which are able to consume simple inorganic compounds (H2 and CO2) for their
metabolism with production methane and acetate, respectively. Such microorganisms could be the best analogues to putative life forms on Mars that could live on simple gaseous substrates. Traces of methane were recently found in martian atmosphere. There is an indication that life could be the most reasonable explanation of its presence. Fe-oxidizing archaeon Ferroplasma acidiphilum inhabits extreme environments on Earth and develops at pH as low as 1. This microorganism is strongly Fe-dependent (Fe is one of the most abundant element on Mars), has a unique enzyme machinery and is recognized as an extremely interesting in respect to industry and biotechnology. The Negev is a rocky desert located in southern Israel. It is dusty mountains interrupted by dry riverbeds and deep craters. The whole Negev region is incredibly arid, receiving very little rain due to its location to the east of the Sahara, high solar radiation and extreme temperatures due to its location. It is a great model system to study rocky desert on Mars. Microbes inhabiting this desert should have specific mechanisms to withstand the extreme environmental conditions. In spite of aeration of the top soil layers, the presence of active strictly anaerobic methanogenic archaea has been shown. Our preliminary experiments revealed a big microbial diversity in the desert samples. Methanogenesis was easily activated after addition of water and flushing the samples with nitrogen. Representatives of Methanosarcina have been detected as predominant methanogens.

A CHAMBER FOR STUDYING PLANETARY ENVIRONMENTS AND ITS APPLICATION TO ASTROBIOLOGY: STABILITY OF LIQUID SALINE WATER ON PRESENT DAY MARS

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An important goal of the exploration of Mars is to determine whether liquid water exists there today or existed in the past. The effect of the environmental parameters at the planet surface, such as radiation, gas pressure, water vapor or temperature, is critical. Perchlorate salts (mostly magnesium and sodium perchlorate) have been detected on Mars’ arctic soil by the Phoenix lander, furthermore chloride salts have been found on the Meridiani and Gusev sites and on widespread deposits on the southern Martian hemisphere. The presence of these salts on the surface is not only relevant because of their ability to lower the freezing point of water, but also because they can absorb water vapor and form a liquid solution (deliquesce). Due to the obvious technical limitations for in-situ planetary exploration, laboratory simulations are one of the most feasible research options to get further in planetary science. We have built a versatile planetary simulation chamber able to reproduce atmosphere and surface temperature for most of the planetary objects. It has been especially developed to make feasible in-situ irradiation and characterization of the sample under study. This chamber achieve total pressure ranging from 5 to 5x10^-9 mbar and partial pressure of the gasses in the chamber can be set with this precision. Desirable atmosphere composition is regulated via a residual gas analyzer with ca ppm precision. Temperature can be set from 4 to 325 K. Irradiation sources up to 5 KV-ions, 5 KV-electrons, Deuterium Ultraviolet (UV) lamp and noble-gas discharge UV are available even at high pressures. Implemented analysis in-situ techniques are UV and infrared spectroscopy. Therefore it allows for recording chemical changes in a given sample upon gas environment, water vapour, temperature and radiation dose. As an experimental case, we have studied stability of liquid brines under martian conditions Our results demonstrate that deliquescent salts such as sodium perchlorate can form liquid water solutions on Mars on locations where salts are in contact with water vapor from the atmosphere or ice reservoirs. We have proven that the deliquescence of sodium perchlorate can form liquid aqueous solutions at temperatures as low as 225 K in a simulated Martian atmosphere at 700 Pa by absorbing water vapor from the atmosphere. We show that the deliquescence of sodium perchlorate is an exothermic reaction that increases the temperature of the sample significantly, and can mitigate freezing at ambient temperatures lower than 225 K. Our results indicate that salty environments make liquid water to be locally and sporadically

**SIMULATING MICROBE-MINERAL INTERACTIONS IN THE SUBSURFACE OF MARS**

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Methane was first observed in the martian atmosphere in 2003. This organic molecule has an expected atmospheric e-folding time of less than 600 years – pointing to a current or recent source of the gas. Several localised methane sources have been postulated, the most likely of which being either the hydration and serpentinization of ultramafic silicate minerals, or the existence of methanogenic life in the planetary sub-surface. Release of the gas by clathrate hydrates has also been suggested; however clathrates are not a source in of themselves, but rather a mechanism of sequestration. If these proved to be linked to the episodic release of methane, an original methane source would still have to be identified. This work is designed to assess the habitability potential of sub-surface Mars by investigating the viability of methanogenic Archaea living on, and interacting with, analogue rocks and minerals. We are quantifying this relationship using the Archaea strains Methanosarcina barkeri and Methanobacterium formicicum as models of such putative martian life. We present here initial results from this study, demonstrating the effect on these microbes of a range of environmental conditions analogous to those thought to exist in the martian subsurface, including pH and water activity.


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ESA and NASA have selected the scientific instruments for their first joint Mars mission. Scheduled for 2016, it will study the chemical makeup of the Martian atmosphere, including methane. Methane was first discovered in Mars-Express spectra and later in earlier telescopic data. No explanation has been given for it until now; the few existing observations can only be explained by a very important fast release and an ultrafast destruction process. They point to underground methane produced from either biological or volcanic sources and sporadically injected in the atmosphere. Five main instruments constitute the payload and are described in the following table; these instruments will confirm the presence of methane and help constrain the models on its origin. The presentation will highlight the payload synergies.

<table>
<thead>
<tr>
<th>Instrument</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Mars Atmospheric Trace Molecule Occultation</strong></td>
<td>An infrared spectrometer to detect very low concentrations of molecular constituents of the atmosphere.</td>
</tr>
<tr>
<td><strong>Spectrometer (MATMOS)</strong></td>
<td></td>
</tr>
<tr>
<td><strong>High-resolution solar occultation and nadir spectrometer (SOIR/NOMAD)</strong></td>
<td>An infrared spectrometer to detect trace constituents in the atmosphere and to map their location on the surface.</td>
</tr>
<tr>
<td><strong>ExoMars Climate Sounder</strong></td>
<td>An infrared radiometer to provide daily global measurements of</td>
</tr>
</tbody>
</table>
dust, water vapour and chemical species in the atmosphere to aid the analysis of the spectrometer data.

**High-resolution Stereo Color Imager (HiSCI)**
A camera to provide 4-colour stereo imaging at 2 m resolution per pixel over an 8.5 km swathe.

**Mars Atmospheric Global Imaging Experiment (MAGIE)**
A wide-angle multi-spectral camera to provide global images in support of the other instruments.

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**THE ULISSE SPACE DATA MANAGEMENT PROJECT AND ASTROBIOLOGY**
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The ULISSE programme has for main purpose the preservation and use of data obtained on the space station and currently recorded by the USOC network in Europe (Users Support and Operation Centres). The data cover physical sciences, life science and environmental parameters relating to the station. They also cover related ground based activities as hospital ground truth, bed rest studies and networks of ground based solar monitoring stations. The data mining aspects of the project have relevance to astrobiology as they relate space physical parameters to biological data. Advancement of the project will be shown together with examples in solar physics prepared for the project demonstrator.

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**THE SURVIVAL OF THE MARTIAN UV CLIMATE-ADAPTED BACILLUS SUBTILIS STRAIN MV01 AFTER EXPOSURE TO SPACE AND MARS CONDITIONS IN THE SPACE EXPERIMENT ADAPT**
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Evolution is the genetic adaptation to changing environmental conditions. It is the result of mutations and natural selection. Solar UV radiation affects life on earth today, and probably has had an even stronger impact on early evolution. The earth's atmosphere at that time was anaerobic and and the UV climate comprises shorter more energy-rich wavelengths resulting from the lack of ozone. The situation on the early Mars might have been comparable. The significance of solar UV radiation as an environmental driving force for the early evolution of life on earth and the adaptation to the current high natural variations of ambient UV levels with latitude, altitude, season, aerosol concentration, clouds, etc. is reflected by the development of different protection mechanisms against the deleterious biological effects of UV radiation. In the space experiment ADAPT the capability of microorganisms to adapt to qualitatively and quantitatively different UV levels like those in space and on Mars were investigated in the ESA facility EXPOSE on the European Columbus module on the ISS. Three highly resistant microorganisms from very distinct terrestrial habitats were selected: *Bacillus subtilis*, a well characterised spore forming bacterium, a natural community of cyanobacteria colonising rocks and a species of halophilic archaea, *Halococcus dombrowskii*. In the experiment ADAPT I the model organism *Bacillus subtilis* was used to test the hypothesis experimentally whether longer-lasting selective pressure by UV radiation of different quality results in a higher UV resistance as well as in a higher resistance against the simultaneous action of further ‘extreme’ environmental factors that exist in space or on Mars like vacuum or cosmic radiation. In addition, the environmental parameters of Mars were simulated in space in EXPOSE by using closed sample carriers with martian atmosphere and pressure and a martian UV climate realised by the use of suitable cut-off filters and
extraterrestrial solar UV radiation. The EXPOSE facility with the experiment ADAPT was launched in February 2008 with STS-122, exposed for 1.5 years on the ISS and brought back to Earth in September 2009 with STS-128. The samples arrived in the PI's lab in December 2009. First results of the survivability of a new UV-adapted *Bacillus subtilis* strain MW01 in space and under martian conditions will be presented.

**ASPICILIA FRUTICULOSA: OPTIMAL BIOLOGICAL MODEL FOR SPACE FLIGHTS, SIMULATION CHAMBER EXPERIMENTS AND INTERPLANETARY TRANSFER TESTS**

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Lichens are one of the most extreme organisms on Earth, due to their capacity to resist and even grow in extreme environments in terms of temperature, UV radiation and desiccation. This resistance is provided by the presence of a thick cortex (upper layer), the production of secondary metabolites with photoprotective effects and, above all, because of their poikilohydric nature. We have selected a vagrant lichen species, *Aspicilia fruticulosa*, whose thalli’s morphology is coralloid with numerous teretes, that grows unattached to the substrate and that is characteristic of arid areas with extreme temperatures and high levels of irradiation. These facts motivated scientists to test its survival capacity in a space flight. After 10 days of exposure in a Low Earth Orbit, a complete recuperation of the vitality of all the samples of *A. fruticulosa* was observed, confirming their high resistance to harsh space conditions. This led us to carry out further experiments in order to test their survivability to Mars surface simulated conditions. Analysis of the results have shown a stable or, in some cases, increased biological activity after exposure to the combination of the physicochemical parameters present in the Mars surface (pressure, temperature, UV irradiation and atmosphere composition) (R. de la Torre et al., Simulation chamber experiments confirm high survival of lichens under Martian conditions, in prep). The experiments performed confirmed that *A. fruticulosa* is one of the most suitable candidates that could survive the second phase of the Lithopanspermia hypothesis (interplanetary transfer of rock inhabiting life by means of meteorites) due to their high resistance, and taking also into account that they reproduce asexually from thalli fragments. A large object impacting on Earth, could hypothetically produce the ejection of rocks from the original planet, lumping some specimens of this widely distributed lichen species. In their natural habitat, they usually develop in cryoturbated soils, at conditions in which they have to face great temperature variations, with daily freezing/thawing cycles, similar to the situation that is supposed to occur in an interplanetary travel. Although the re-entry step to a planetary atmosphere by a biological test system (the lichen species *Rhizocarpon geographicum* tested in ESA’s last Foton M3 mission (2007) with the Lithopanspermia-Stone experiment) demonstrated low survival chances, perhaps they could be increased by a reduced fragment of an *A. fruticulosa* lichen sample embedded in the mass of an “hypothetical meteorite”.

References:


BACILLUS PUMILUS SPORES HITCHHIKING TO ISS – SURVIVABILITY AFTER EXPOSURE TO SPACE AND SIMULATED MARS CONDITIONS

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Space is considered an extreme environment due to the intense galactic and solar radiation, extreme variations in temperature, microgravity, and high vacuum. When microorganisms are not protected against radiation, either by a thin layer of dust or the geometry of the spacecraft, the prevalent conditions in space are extremely harsh for their survival and subsequent growth. Although several laboratory space simulation experiments revealed that microbes can withstand extreme environmental conditions, direct exposure to “real space” will yield appropriate empirical data on their survival mechanisms at the molecular level. Spores of the well studied and completely sequenced Bacillus pumilus SAFR-032 strain, isolated from a spacecraft assembly environment, were flown to the International Space Station (ISS) as part of DLR’s EXPOSE-E experiment PROTECT and exposed to a variety of space conditions using the European Technology Exposure Platform and Experiment Facility (EuTEF) for 18 months (Feb, 2008 to Sep, 2009). The results of the ground simulation experiments showed that the desiccated spores survived full Martian UV (200 - 400 nm) exposure (87 hours; 30 W m²) and exhibited only a 2-log reduction in viability. After exposure in the EuTEF facility under dark space conditions, SAFR-032 spores showed 10-40% survivability whereas spores kept under dark simulated Mars atmosphere showed a survival rate of 85-100%. In contrast, when space UV (>110nm) was exerted on SAFR-032 spores for the same time period and conditions using the EuTEF, a ~7-log reduction in viability was observed but few colonies were isolated. This might be due to the shadowing effect in which some of the spores found the faulty spots of the aluminum, and thus a place to hide, and survived the UV exposure. Representative space-surviving spores (Space UV, Space Dark, Mars UV, and Mars Dark) exhibiting enhanced UV resistance (4k Jm²; UVC) when compared to ground-control spores (2K Jm²; UVC) were selected to characterize genetic and proteomic changes. Comparative two-dimensional gel electrophoresis and mass spectrophotometry protein identification revealed compelling evidence of marked differences in protein profile between ground-control and space-surviving spores. The preliminary results showed notable elevated changes in the superoxide dismutase concentration in the “real space” UV exposed spores compared to their ground control counterparts. The data generated are important to assess the probability and mechanisms of microbial survival, microbial contaminants of risk for forward contamination, in situ life detection, and to safeguard the integrity of sample return missions. This study provided unique insights into the survivability and possible proliferation of microbes in extreme extraterrestrial environments. Acknowledgments. The research described in this publication was carried out at JPL, California Inst. Tech., under a contract with NASA.

PRESERVATION OF LIFE PROCESS (PHOTOSYNTHESIS) OF LICHENS

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Survival capacity of lichens, to LEO (Low Earth Orbit) space conditions, have been tested at three space missions of ESA the first two were short missions on board of ESA’s facility Biopan of the Foton satellite, and the third one, a long-term mission, on the Expose facility of the ISS. Aim was to experimentally test the likelihood of interplanetary transfer of rock inhabiting life by means of meteorites, a step belonging to the hypothesis of lithopanspermia, which argues that impact expelled rocks from a planet’s surface serve as vehicles for spreading living material from one planet or solar system to another (Nicholson et al. 2000, Cockell 2008). The first two experiments, Lichens (Biopan-5, Foton M2 mission, May 2005) and Lithopanspermia (Biopan-6, Foton-M3 satellite, 14.09.07), allowed, for the first time, the demonstration- and intercomparison of the high survival capacity of eukaryotic- (and prokaryotic) symbiotic organisms in space (the epilithic lichen species R. geographicum and X. elegans, endoevaporitic microbial communities, cyanobacterial akinetes, and a vagrant lichen species, Aspicilia fruticulosa). With the exposure to space- and Mars environment (full spectrum of solar extraterrestrial electromagnetic radiation or selected wavelength ranges, space vacuum, cosmic radiation and extreme temperatures), we checked the viability-, resistance- and adaptation mechanisms of lichens and their symbionts (mycobiont/ photobiont). In this work we expose the results obtained after flight, that show the recovery of the biological activity (chlorophyll a- fluorescence) of the epilithic- and vagrant lichen species (de la Torre et al. 2007; Sancho et al., 2007), very high after short missions, and clearly decreased, after a long 18 months exposure, an important knowledge that lead us to learn, that symbiotic eukaryotic organisms adapted to tolerate extreme conditions on our planet, like epilithic- and vagrant lichens, have developed optimal resistance- and adaptation mechanisms that allowed them to preserve their life processes (photosynthesis, growth and germination capacity) after space- and Mars exposure, due to their symbiotic nature. They could resist an interplanetary travel through space, regarding them as potential interplanetary propagules.
Theme 4: Solar system exploration

**LIFE DETECTION STRATEGY FOR JOVIANICY MOONS: LESSONS FROM SUBGLACIAL LAKE VOSTOK EXPLORATION**

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The objective was to estimate the microbial abundance and diversity in accretion ice originating from the subglacial Lake Vostok buried beneath 4-km thick East Antarctic ice sheet with the ultimate goal to discover alien life in this extreme icy environment. The DNA study constrained by Ancient DNA research criteria was used as a main approach. The flow cytometry was implemented in cell enumerating. At present the drill at Vostok reached the depth ~3650m (within the 210m accretion ice layer, about 100m towards the ice-water boundary).

As a result, the molecular microbiology study showed that the ice until depth 3659 m contains the very low microbial biomass [1]. The only ice containing mica-clay inclusions allowed the detection of unusual gas content and recovery of few bacterial phylotypes all passing contaminant controls but not fitting groups expecting to discover. The latter included the well-known chemolithoautotrophic thermophile *Hydrogenophilus thermoluteolus* ([β-Proteobacteria]) [1, 3] along with unclassified uncultured bacterium of OP11 Candidate Division (91% similarity with closest relative). In contrast, the deeper accretion ice with no sediments present and gas content close to detection limit gave no reliable signals. In addition, accretion ice contains dissolved organic carbon (DOC) as low as <20 ppbc, implying ultra-oligotrophic conditions and restricting biota to being chemoautotrophic.

Thus, the search for life in the Lake Vostok is constrained by a high chance of forward-contamination and the subglacial Lake Vostok may be viewed as the only extremely clean giant aquatic system on the Earth providing a unique test area for searching for tiny life indices (assumed to be DNA-based) on icy worlds such as Jovian moon Europa.

As a conclusion, the life detection strategy for (sub)glacial environments existing on Earth or the Jovian moon Europa (for a hypothetical robotized in-situ analyser) should be based on: (i) applying stringent ice sample (and tools to work on) decontamination procedures in clean conditions to meet trace chemistry and Ancient DNA analysis standards, (ii) certification of the various environments in contact with the ice samples for biological content (establishment of contaminant library), (iii) implementation of appropriate methods to obtain a signal at as low as possible a detection level, (iv) verification of findings through their possible metabolic profiles as deduced from physical/chemical features of the environment under consideration, (v) replication of findings at an independent laboratory (or experiment) within the framework of several specialized laboratories.

References:

**TITAN ATMOSPHERIC COMPOSITION FROM UVIS DATA ANALYSIS**

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Cassini-Huygens is the NASA-ESA-ASI mission to explore the Saturnian system. One particularly interesting object is Titan, Saturn’s largest moon and the only one in the Solar System known to have
a thick N2/CH4, planet-like atmosphere. This atmosphere is long ago known to be rich in organic compounds of high interest for astrobiology. Together with other 11 instruments the Cassini orbiter has the Ultra Violet Imaging Spectrometer (UVIS), a valuable tool that, among other capabilities, allows studying composition, element distribution and temperatures in Titan’s upper atmosphere. Star occultation by Titan’s atmosphere is a convenient technique to retrieve altitude profiles of absorbing compounds. Cassini fly-bys provide excellent opportunities for this. The quality of the result is highly dependent on the resolution of the measured transmission spectra, and on the available molecular absorption cross section data for the atmosphere’s component species used in the retrieval (Ferradaz et al., 2008). In this work, transmission spectra for different altitudes in Titan’s atmosphere are obtained from UVIS data. The data, in the form of spatial spectral cubes, corresponds to star occultation events. The data products include those of the December 2004 Titan fly-by (Shemansky et al., 2005). The transmission spectra are used together with recent experimental measurements of absorption cross sections of the relevant molecules (Ferradaz et al., 2008) to determine the presence and distribution of carbonaceous compounds in Titan’s atmosphere. The modelled atmosphere used for the retrieval includes CH4, C2H2, HCN, C2H4, C2H6, C4H2, HC3N and C2N2. It is expected that the UVIS spectral resolution and absorption cross section, improved with respect to previous instruments and experimental data respectively (Vervak et al., 2004), will result in a very reliable identification of species densities from the transmission spectra. The availability of cross section information for different temperatures will also improve the reliability of the retrieved abundances. This analysis will be compared to available photochemical models, which will help to improve the comprehension of Titan’s atmospheric organic chemistry.

DETECTING MOLECULAR SIGNATURES OF LIFE ON MARS: THE LIFE MARKER CHIP (LMC) INSTRUMENT

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In recent years, the rise of interest in planetary exploration and the emergence of Astrobiology as a promising field of research have lead to a number of programmes aiming to develop sensitive instruments for the detection of the molecular signatures of life in extreme environments. An antibody assay-based life detection instrument, the Life Marker Chip (LMC), is currently under development by a UK-lead international consortium for the European Space Agency’s (ESA) ExoMars rover. This forms part of the joint ESA/NASA Mars exploration programme with the ExoMars Rover currently scheduled for launch in 2018. The organic molecules targeted for Life detection by the LMC are based on an assumption of “Earth-like” Life on Mars – extinct and/or extant. The molecular targets for the LMC have been chosen to represent markers of extinct Life, extant Life, abiotic chemistry (e.g. of meteoritic origin) and mission-borne Earth contamination. The LMC incorporates integrated liquid sample extraction and processing for dry Martian samples, which will be collected from up to 2m below the surface of Mars, where organic molecules, if present, are expected to be better preserved. The core technology of the LMC is a combination of optical evanescent waveguides, micro-fluidics, immuno-microarrays with fluorescent labels and CCD detector readout. Phage display recombinant antibody technology has been employed in order to acquire antibodies against a number of the LMC target molecules. The LMC hardware is currently in a breadboard phase of development. The recombinant antibody development for LMC targets is an on-going project, and testing of Earth-analogue Martian samples has been initiated.
ACCRETION OF INTERPLANETARY AND INTERSTELLAR DUST PARTICLES AS AN IMPORTANT SOURCE OF THE PREBIOTIC ORGANIC ON THE EARLY MARS

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Accretion of interplanetary and interstellar dust particles is studying as an possible source of the prebiotic organic on the early Mars. Modeling studies of the early terrestrial atmospheres found that the upper atmosphere of early Noachian Mars was hot and highly expanded CO₂ atmosphere under the strong EUV flux from the young Sun [1]. Besides that, direct measurements reveal intact organic macromolecules in interplanetary and interstellar dust particles collected in the stratosphere and polar ices of the Earth [2, 3]. Delivery of cosmic dust organic take place on Mars too. We show increase in the cross-section of dust particles capture by ancient atmosphere vs present Mars atmosphere. Also, atmospheric entry heating of dust particles in early extended atmosphere is lower in comparison with present atmosphere. Therefore accretion of interplanetary dust particles could have been most important source of the prebiotic organic on the ancient terrestrial planets.

References:

GAS ANALYTICAL COMPLEX FOR INVESTIGATION OF VOLATILE COMPOUNDS ON PHOBOS, MOON AND EUROPA

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The possibility of prebiotic synthesis of organic molecules is defined by a number of physicochemical conditions in the system, such as the presence of appropriate atmosphere, availability of water, sources of free energy, combination «reactor-refrigerator» etc. Various experiments on prebiotic synthesis have been successfully carried out in modeled earthly conditions. But before present time no organic substances have been found in investigated terrestrial group planets and satellites. Giant planets system indicates the presence of organic compounds. Nevertheless, no life forms were detected in the Solar system other than on Earth and other world can be considered as prebiotic. Therefore visits to a hypothetic «prebiotic worlds» such as planets, asteroids and bodies without atmosphere (such as satellites) may give interesting information about prebiotic synthesis in specific space conditions. Russia is planning several planetary missions with landers in near future: the Phobos Sample Return mission (2011 year), the Moon-Globe (2012 year), the Moon-Resource (2013 year), and the Europa-Lander (approx. 2020 year). The gas analytical complex is among the main instruments in the planning payload of those spacecrafts. This complex consists of the thermo differential analyzer, the gas chromatograph, and the mass-spectrometer. The complex provides release of volatile components from soil or ice, concentration in cooling traps with subsequent thermal desorption, chromatographic separation and detection of volatile compounds including organic compounds. Special attention is paid to research of ice shell of Jupiter’s satellite Europa. The peculiarity of Europa comes from the presence of hypothetic water ocean under the ice shell which can be favourable to the habitability. Synthesis of organic compounds on Europa can be due to various energy sources: solar UV- radiation, Jupiter radiation belts, deep- ocean volcanism,
etc. The combination of «reactor-refrigerator» is also realised at this icy satellite. Gas analytical complex is aimed to investigate the presence of organic substances in such «prebiotic worlds».

THE DEVELOPMENT OF INSTRUMENTATION FOR MARS EXPLORATION AT THE SPANISH CENTRE FOR ASTROBIOLOGY (CAB)

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The exploration of Mars, looking for traces of water or potential habitability conditions is essential for Astrobiology. At CAB in Madrid we have developed a series of advanced instruments for the characterization of the Martian surface conditions and its meteorological content. The REMS instrument is an environmental monitoring device for the MSL rover of NASA to be launched in 2011. It measures speed and direction of the wind, surface and air temperature, pressure, relative humidity and the incidence of ultraviolet radiation. The second experiment is a Raman spectrometer for the analysis of the content of subsurface samples extracted by the drilling mechanism of the Exomars rover, currently being developed by ESA for launch in 2018. Meanwhile, other experiments are being studied, like a proposal for a nadir-scanning thermal infrared radiometer to be embarked on the Exomars Orbiter in 2016 and the identification of complex organic compounds via microarrays of antibodies to be presented to the next available flight opportunity.

EVOLUTION OF THE MARTIAN SURFACE PRESSURE AFTER THE NOACHIAN

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Lower energetic neutral atoms which are produced due to photochemically reactions in planetary thermospheres play an important role for the escape of atmospheric species at lower mass planets, like Mars. The newly photochemically generated energetic neutral atoms, O, C, N, and H are traced from their point of origin up to the exobase or beyond, by using a 3D Monte-Carlo model and the kinetics and transport characteristics of these particles are determined. The simulation considers the collision of the suprathermal particles with the background gas, energy transfer, and the tracing of secondary and cascaded hot atoms, which are generated in collisions of the hot particles with ambient constituents. A non-linear electron dissociative recombination coefficient as well as energy and mass dependent collision cross sections and their corresponding scattering angles (forward scattering) are also taken into account. The exosphere density is obtained from the corresponding energy density and angular distribution at the exobase altitude by using a test particle model which traces the ballistic trajectories of hot atoms in the exosphere. In this model we consider inelastic and quenching collisions between the traced hot particle and the ambient neutral atmosphere as well as differential cross sections to determine the scattering angle in the collisions. We also include rotational and vibrational excitation energies for the calculation of the initial energy of the produced hot oxygen, carbon, nitrogen, and hydrogen atoms. With the simulation of the loss rates for O, C, N, and H over the past, starting from the end of the Noachian Epoch up to now, the loss of the CO2 atmosphere during the history can be calculated. With these losses one can estimate the evolution of the surface
pressure and its influence to the climate conditions and to the planetary habitability caused by hot atom escape.

THE SEASONAL BEHAVIOR OF ICE IN CRATERS AT THE MARTIAN NORTHERN POLAR REGION

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Introduction: 87 Martian craters in the Northern Polar Region have been monitored poleward of 60° in latitude, in order to better understand how the Martian weather conditions affect water and carbon dioxide ice annually in craters. Each crater has been examined during varying solar longitude, during which the amount of ice has shown both expected and unexpected seasonal variations. CTX and HiRISE: The Context Camera (CTX) on the Mars Reconnaissance Orbiter (MRO) is a camera providing black and white context images of the Martian surface, with spatial resolution of 6m/pixel [1]. These CTX images are used as a complement for the High spatial Resolution Imaging Science Experiment (HiRISE) camera, with 0.25 to 1.3 m/pixel images [2]. Crater mapping: Every crater has a set of images with data we have used to create a database, called Information on Craters in the Martian Northern Polar Region. The craters chosen to be investigated are either named or have a diameter of at least 10km. In addition to image observation the amount of ice (none, less than 50%, more than 50% and full), unusual features, crater data and a description of the area are recorded.

Analysis: The closer the crater is to the NPRC, located poleward of 80° in latitude, the more ice it contains and is preserved more or less the whole Martian year. Korolev is one of these craters, located at 73° in latitude. Two craters, Lomonosov and an unnamed crater, located on the eastern and the western side of the NPR, at the same latitude of 65N and during the same Martian season, shows a clear contrast in the observed amount of ice covering it. Sevel crater, located at 79° in latitude, is another crater with abnormal behavior as it is fully covered with ice during the summer.

Conclusions: Anomalous seasonal behavior of ice in craters could be explained by the type of ice. Latitudinal variations in insulation which drives the atmospheric circulation of water, carbon dioxide and airborne dust [3] affect the constituents of the ice. This in turn determines the characteristics and melting process of the ice [4]. Surface albedo, affected by the size of the ice grain [5] and dust [6], is an important factor as it determines the amount of absorbed energy. Our observations suggest Korolev experiences a periodic oscillation in the amount of ice coverage during the spring season, in Mars Year 29. Most likely factors affecting the periodic oscillation are the length of day and night, distance to the sun, the solar angle, latitude, altitude and seasonal storms. Craters below Korolev and above Sevel, in latitude, do not show the same type of periodic oscillations. More images have to be taken in high resolution with HiRISE, over different seasons, to monitor the change of surface albedo in craters over time.

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TOWARDS A EUROPEAN VISION OF SPACE EXPLORATION

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Space exploration is a rapidly evolving global venture. Fourteen space-faring nations through their space agencies – including ESA plus several European member states – have agreed on a coordinated approach, the “Global Exploration Strategy” (2007). In view of this increasing interest in space exploration at the global scale, the Space Advisory Group of the European Commission has evaluated the situation in Europe with regard to its potentials to participate in this ambitious global enterprise. Aspects of science, technology, environment and safety, society, spin-offs and international cooperation have been considered. The group has come to the conclusion that Europe possesses sufficient key technologies and scientific expertise to play a major role in international space exploration. It has been recommended that the EU takes a central role to ensure the success of future European space exploration, not only to give a clear political signal for the way forward but also to ensure an appropriate financial framework. Europe will thus embrace the spirit of the European Space Policy and contribute to the knowledge-based society by investing significantly in space-based science and technology and thereby playing a strong role in international space exploration (Horneck et al. 2010)

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EFFECT OF IMPACTS ON THE EVOLUTION OF MARTIAN ATMOSPHERE DURING THE LATE NOACHIAN EPOCH

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It is likely that Mars had a denser atmosphere with sufficient greenhouse gases to sustain the presence of stable liquid water at the surface during the late Noachian epoch. This denser atmosphere was lost since that time via different escape mechanisms including ion erosion, sputtering and impacts. In the present study, we investigate the effect of impacts on atmospheric evolution of Mars using a semi-analytical model that that takes into account the numerical results of several impact simulations. We show that asteroids and comets impacts affect the atmospheric evolution not only by causing atmospheric erosion but also by delivering material and volatiles to the planet. In our study we consider also the possibility of an increased impactor flux in during the late Noachian epoch.

OUTGASSING AND LOSS OF THE MARTIAN ATMOSPHERE DURING THE NOACHIAN EPOCH

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From spacecraft observations there are geological evidences suggesting that early Mars was more wet and warmer during the late Noachian epoch. However, such climate conditions require most likely a denser CO2 atmosphere. Astrophysical observations of young solar like stars indicate that the young Sun was very active in the X-ray and stronger in EUV radiation. In recent model simulations it was shown that this environment may have caused a problem for the stability of the early CO2 atmosphere during the Noachian. The high solar EUV flux would have dissociated CO2 molecules in the thermosphere so that thermospheric cooling was reduced. The enhanced heating expanded the atmosphere to higher altitudes and most likely even above the early martian magnetosphere. In this work we study various outgassing scenarios of CO2 from the interior of the planet and combine the outgassing rates with atmospheric evolution scenarios including impacts during the heavy bombardment, thermal escape and non-thermal solar wind induced ion erosion during the first Gyr of the planet’s origin. We investigate and present scenarios where the CO2 outgassing rate may have been large enough, at some time period after the planet’s origin, so that the loss rates were lower, compared to the outgassing rates and a denser CO2 atmosphere could have build up around 4 Gyr ago. This denser atmosphere was lost since that time via ion erosion, sputtering and hot atom escape. Finally we address astrobiological implications for the time, when life may have originated on early Mars.

**METHODOLOGY AND DEVICES FOR DETERMINATION OF SIGNS OF LIFE UNDER THE ANALYSIS OF A MICROBE BIOMASS IN ICES OF EUROPE**

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Providing the presence of simplest forms of life in the ocean of Europe, the biomass of these organisms that have remained in an ice matrix is supposed to be detected also in the surface layer of ice. For the investigation of signs of life is proposed the measuring complex of two time-of-flight mass spectrometers (TOF MS): laser and gas, and the system for the biomass extraction from the water obtained from ice sample.

Signs of life can be detected by measurements of:
- element composition of the biomass sample with the help of laser TOF MS, by ratio of mass peaks of C, O, N, H, also K, Ca, P, S and some microelements;
- masses of molecular ions obtained from the biomass sample after its thermal evaporation and ionization in the gas phase by electron impact, with the help of gas TOF MS;
- molecular mass of secondary ions located in the ice matrix, emitted under the influence of the primary energetic particles beams on the surface of Europe in the processes of similar fast atom bombardment, with the help of gas TOF MS operating in a mode of external ions registration.

The joint comparative analysis of these results will give the information about the possibility of presence of life on Europe and its similarity with Earth form. The instrument complex can be created after considerable modernization of the new generation onboard instruments LASMA and MANAGA developed for Phobos – Soil Mission. Onboard system of biomass extraction will be created for the first time.
NUCLEAR INSTRUMENTS FOR TESTING THE PRESENCE OF WATER IN THE SUBSURFACE OF MARS, MOON AND OTHER CELESTIAL BODIES OF THE SOLAR SYSTEM

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Nuclear Instruments and experimental data will be presented for testing the presence of water in the subsurface of Mars, Moon and other celestial bodies.

COSMIC RAY IMPACT ON SURVIVAL OF TRACES OF LIFE IN MARITIAN SUBSURFACE LAYER

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A problem connected with a search for life or traces of life on Mars is studied, namely, the radiation sterilization of the planet surface by cosmic rays. We simulated the process of radiation dose accumulation in subsurface layer of Mars using the modern GEANT 4 procedure for simulation of ionization by cosmic rays. The GEANT 4 takes into account in a most contemporary manner the processes of nuclear and electromagnetic interactions of primary and secondary particles with compound media. Physical conditions on Martian surface now and its variations in time as well as effect of cosmic rays variations are analyzed. We show that the average dose intensity in the upper meters of Martian soil is not dangerous for living microorganisms. However, sterilization of microorganisms in the dormancy state would occur in the upper 20 g·cm⁻² in less then 30 thousand years. The dormant life would be killed in the layer down to about 700 g·cm⁻² by the galactic cosmic rays in a time period less then 2 × 10⁶ yr. Destructive effect of cosmic radiation is also able to decrease significantly the mean atomic mass of hypothetical Martian fossil organic macromolecules and¹³C/¹²C ratio in subsurface layer. We make conclusions about locations of most favorite places for the search for life or its traces in the Martian subsurface layer.

PLANETARY CYCLE OF CLIMATE AND LIFE

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Planetary cycle of life[1] Every planet are subject to planetary cycle of climate and life induced by planet-central_star(+gas-giant) interaction. Planetary crust is thin at the beginning of planet formation, volcano activity (VA) powerful, frequently. When volcanic activity is on the decrease on a planet - the life is on the decrease too [2]. VA is a destroying force of bio-life. «Nuclear winter», the biblical Flood, clouds disappear intensity of life grow - quantities of populations and individuals - ‘nuclear spring’. 2 extremes - nuclear winter - Mars nuclear summer - Venus Scenario of events leading to life’s crisis. Long solar activity. Pause. Increasing VA-volcano activity. Local -in time-decreasing of temperature. Explosive increasing VA. Dust, decreasing of temperature. Life down. Pause. VA down. Clouds, dust has been reduced TITAN [3]. We may presume that we watch on Titan ice age or nuclear winter. This ice age take place over planetary scale. The reason of this nuclear winter is volcano activity. The reason of volcano activity is Titan-Sun (+Saturn) interaction.
Titan has already atmosphere, so it has volcanism. One may find life on Titan around volcanoes like in oceanic depth around Smokers[4].

References:

JAPAN ASTROBIOLOGY MARS PROJECT (JAMP): SEARCH FOR METHANE-OXIDIZING MICROBES ON MARS SURFACE
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The liquid water is considered to be a critical factor for life. Gibbs free energy is another factor that should be counted to sustain life for long duration. The Gibbs free energy is obtained by reaction between reductant and oxidant, or from any other non-equilibrium state of matter. As an example, aerobic organisms use carbohydrate and oxygen for getting Gibbs free energy. Many types of chemoaotrophic mechanisms are known for the process as well. On Mars surface, methane and oxidative compound such as ferric oxide or sulfate are found, and they can be source of Gibbs free energy. Iron-dependent methane oxidizing bacteria was found in marine environment on Earth [1]. This finding suggests possible presence of methane-oxidizing bacteria on Mars surface, if local thermal environment and other resources permit proliferation and metabolism of the bacteria during limited portion of time period. Our project aims to search for the methane-oxidizing microbes on Mars surface. Martian soil will be sampled from a depth of about 5 or 10 cm below the surface, where organisms are supposed to be protected from harsh hyper-oxidative environment of Mars surface. Small particles less than 0.1 mm are sieved from the sample, before transferred to analysis section by a micro-actuator. The particles are stained by cocktail of fluorescent reagents, and examined with a fluorescent microscope. Combination of fluorescent dyes is selected to identify life forms from the soil sample. Intercalating fluorescent dye such as SYBR Green is used to detect genetic compounds such as DNA. Membrane specific dye or the combination of dyes is used to detect membrane surrounding the “cell”. Substrate dye that emits fluorescence upon cleavage by the catalytic reaction is used to detect the catalytic activity of the “cell”. A combination of staining reagents is chosen based on the definition of life. DNA or genetic material is required for “replication” of life form. Membrane separating cell from ambient leads to identification of “individual”. Catalytic reaction of enzymes drives “metabolism”. The combination is useful also for detecting pre-biotic organic material as well as remnant of ancient life. Hydrolysis of the polymers in the “cell” followed by HPLC or soft ionization MS for amino acid analysis is effective in examining whether Martian life is identical or different from terrestrial life. The number and type of the amino
acids as well as chirality will be analyzed to distinguish if the polymers are contamination made by Earth-related life form.

References:

**VISUALIZATION OF SOIL MICROORGANISMS IN EXTREME ENVIRONMENTS BY FLUORESCENT MICROSCOPY**

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Fluorescent microscopy is a potentially powerful tool to detect extraterrestrial life. It is high sensitive and visualizes life form directly. Much kind of fluorescent dyes is commercially available. For example, DAPI and SYBR Green detect nucleic acids. ANS and FM1-43 detect cell membranes. CFDA and CFDA-AM detect enzyme activities. By the combination of these dyes, it would be possible to detect various types of life form. But this method has problems in the application of field samples, especially soils. A lot of dyes bind to mineral particles nonspecifically and also some dyes show high background fluorescence. In this study we have used quenching compounds to absorb extracellular fluorescence and successfully visualized soil microorganisms in Antarctica, high mountains and so on. It is discussed how to use fluorescent microscopy to detect life on Mars.

**EUROPA LANDER MISSION**

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An international effort dedicated to science exploration of Jupiter system planned by ESA and NASA in the beginning of next decade includes. Europa Lander mission is planned by Russian Space Agency and the Academy of Science for in-depth science investigation of Europa. To be launched in parallel with EJSM this mission will include a small telecommunication and science orbiter and the surface element: Europa Lander. In-situ methods on the lander would provide the only direct possibility to assess environmental conditions, and to perform the search for signatures of life. A critical advantage of such in situ analysis is the possibility to enhance concentration and detection limits and to provide ground truth for orbital measurements. The science mission of the lander is biological, geophysical, chemical, and environmental characterizations of the Europa surface. Remote investigations from the orbit around Europa would not be sufficient to address fully the astrobiology, geodesy, and geology goals. The science objectives of the planned mission, the synergy between the Europa Lander and EJSM mission elements, and a brief description of the Laplace-Europa Lander mission will be presented.
This study is devoted to description of the possible results that might accompany collisions of natural cosmic bodies with both a planet’s atmosphere and surface. The methodology of the classification is based on the analytical solution of differential equations of meteor physics. These equations characterize the body’s trajectory in the atmosphere, namely, the dependences of the body’s velocity and mass on the flight altitude. The solution depends on two dimensionless parameters defining the drag rate and altitude, and the role of the meteoroid’s mass loss when it moves in an atmosphere. The action of the collisions on the planet’s surface essentially depends on values of these two parameters. Additionally, we formulate recommendations for further studies of the important problem related to the interaction of cosmic bodies with planet atmospheres.

The search for extinct or extant life on Mars is focussing on the detection of diminutive traces of biological origin, such as cell wall fragments, biologically precipitated minerals or non-stochastic distributions of life-specific amino acids. Hence, one of the major concerns for the exploration of Mars is the inadvert contamination of soil samples with bioloads introduced by both robotic and human explorers leading to a false positive detection of "life". In order to investigate these contamination vectors, the Austrian Space Forum has initiated a multiyear interdisciplinary research programme ("PolAres") to study human-robotic interaction in a set of field tests culminating in an arctic expedition. The astrobiological focus of these field simulations is to assess bioload transfer issues during a human Mars surface sojourn. In a realistic arctic simulation a carefully selected and trained crew will operate spacesuit simulators, a Mars analogue rover, a custom designed drilling rig and other complementary equipment. They will study subsurface ice deposits with geophysical methods, establish the drilling infrastructure and acquire a pristine subsurface soil core for sample-return. As the High Arctic does not offer a sterile environment, fluorescent microspherules have been selected as a microbiological proxy to emulate diminutive traces of life. One of the core elements of this integrated mission simulation is a set of planetary surface spacesuits ("Aouda"). This spacesuit prototype is an advanced simulator providing all major limitations a real spacesuit would have on Mars whilst integrating a suit infrastructure which provides a high situational awareness. Aouda is based upon a Hard-Upper-Torso design using an semirigid exoskeleton to mimic the pressure suits movement restrictions modelled after various spacesuit flight models w.r.t. its flexion angle vs torque behaviour. Due to its adjustability, various pressure regimes can be simulated. Aouda has been optimized to reduce the amount of biological forward and backward contamination in order to study planetary protection issues during a planetary sojourn. We present the design and engineering challenges as well as the first qualification and benchmarking tests of the Aouda prototype which shall then be evolved into four field units. We also report on the first laboratory tests using these proxies on various surfaces and their adhesive behaviour compared to real biological substrata under
simulated Martian conditions and explores its potential to ensure a pristine sample acquisition for future missions.

HABITABILITY OF EXOPLANET MOONS

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Most of the over 460 known (June 2010) exoplanets are gas giants larger than Neptune. They are unable to harbour life as we know it. This is because of their gaseous structure and also because most of them are located closer to or further from their host star than the local Habitable Zone (HZ).

The giant planets in our own Solar System have a large variety of satellites around them. The largest satellites or moons vary from spheres consisting of rock and ice and to volcanically very active and almost molten objects. As exomoons have not yet been found we have to consider the natural satellite families in our Solar System to be an exemplary representation of satellites in general, from which we will expand our calculations.

The orbital dynamic behaviour of moons around their host planets are examined through numerical simulations. These are based on current theories of moon formation and include gravitational effects of other planets. We will consider orbit circularisation and tidal locking. The formation mechanism and hence also the dynamics are important factors on the way to understanding the possibilities for suitable environments on the surfaces of exomoons.

As a moon is constantly under the influence of its central planet the physical environment on the moon may be very different from the one expected from the star-planet distance only. Especially, the local chemical composition (i.e internal and surface composition and atmospheric chemistry) and the effects of tidal forces reflect the presence of the planet. These conditions lead us to the concept of the 'satellite-HZ', which is separate from the concept of normal HZ.

We discuss various dynamical and physical limits on the habitability of moons around known exoplanets. We will present also limits for the 'satellite-Hz' Some observational suggestions are also given.

THE INTERIOR OF TERRESTRIAL-LIKE EXOPLANETS

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More than 450 extrasolar planets have been discovered in the last twenty years. Even though most of them can be characterized as gas planets, also some examples for terrestrial-like exoplanets are known. To acquire knowledge on the internal structure of these superearths in most cases only observational data on their masses and volumes are available and can be used for further analyses. For some exoplanets (e.g. Corot 7-b, Leitzinger et al., 2009) also the scenario that this planet is the remaining core of a previous gas planet cannot be ruled out, but a model for its interior can be used to give a further argument against this hypothesis and to show that it is more reasonable that Corot 7-b was a terrestrial planet throughout its whole lifetime. We have used an uncompressed density model and defined two internal layers (core and mantle) for the exoplanets of interest to be able to calculate
the sizes of their cores and mantles. Different model scenarios with respect to variations in the Fe, Ni, S contents in the core of the planets as well as different scenarios for their mantle compositions (also with respect to the spectral classes of the central stars) have been implemented. We will present a list of extrasolar terrestrial-like planets, their bulk and uncompressed densities and resulting core diameters and mantle dimensions. In the context of Corot 7-b we have investigated several different density scenarios and it is more likely that only for the scenario that the atmosphere is very “ragged”, it could be possible that Corot 7-b was a gas planet in its early history.

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SOLVENTS OF INTEREST FOR EXOTIC LIFE ON PLANETARY BODIES IN LIFE SUPPORTING ZONES AROUND MAIN SEQUENCE STARS
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Life on Earth shows specific characteristics regarding its biochemistry, whereby two attributes of Earth-type life are: 1) the use of water as solvent and 2) that the molecules of life can pass through various transformations (e.g. metabolisms). This was suggested to be a consequence of the chemical characteristics of C, O, H, N, S and P (Baross et al. 2007). The concept of exotic life generalizes at least these two properties: it is not necessarily based on water as a solvent and a potential metabolism has not to be based on the chemical characteristics of these elements (Leitner et al., 2010). As a consequence the concept of the life supporting zone was introduced expanding the traditional habitable zone (Kasting et al., 1993), which reflected a geocentric way of thinking in questions related to the habitability of a planet. The life supporting zone is composed of additional habitable zones with different solvents. These zones can merge and form together a region around a central star where exotic life could be possible. However, one question still remains unanswered: what are the possible building blocks and solvents for exotic life? Miller-Urey experiments will be performed to find out which solvents are suitable for the formation of peptides and other macromolecules that could conceivably be utilized by exotic life. In this context we will not only investigate the question of non-standard solvents, but will also perform experiments with water-ammonia composites to find constraints for the amount of ammonia which can be dissolved in the oceans of the icy moons of the outer Solar System (Hussmann et al., 2006), but still allow life as we know it. Earlier Miller-Urey experiments have shown that mainly amino acids or predecessor molecules will result if water is involved as a solvent (Rode et al., 2007; Plankensteiner et al., 2007), but if water is not part of the chemical reaction, it is unknown which macromolecules may form.

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PAENIBACILLUS- A TYPICAL CLEAN ROOM CONTAMINANT POSING A THREAT TO PLANETARY PROTECTION?

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The multiresistance properties of spore-forming microorganisms make them ideal candidates for the survival of space flights. Carried to planets of significant interest concerning biological and chemical evolution, these microbes could cause false positives when searching for extraterrestrial life. Therefore, the sterility of a spacecraft is a desirable goal, but the sensitive hardware does not allow an overall sterilization after assembly. Disinfection via chemicals or other sterilization procedures that do not harm the spacecraft and its instruments are not able to kill all bacterial spores. Innumerable cultivation assays from Viking and other spacecraft have proven the presence of a broad variety of spore-forming microorganisms on spacecraft related surfaces. Based on the first results from the 70’s, current space agency protocols concentrate on the detection of aerobic, mesophilic bacterial spores for the estimation of the overall bioload. In particular representatives of the genus Bacillus have been regarded as one of the most harmful and resistant spore-forming microorganisms for planetary protection related issues. Nevertheless, other spore-forming microbes have been isolated from spacecraft assembly facilities. Especially Paenibacillus strains were recurrently obtained during biodiversity studies of such clean rooms all over the world. These “almost” bacilli (L. adv. paene, almost) have not been considered so far as a significant contamination source in planetary protection relevant environments, although they seem to be at least as resistant as the closely related Bacillus. In a very recent study, several Paenibacillus strains have been isolated from European spacecraft assembly clean rooms, many representing novel species. Our studies have shown, that most of the isolated strains are able to fix nitrogen, grow at low temperatures, can resist immense temperature and pH shifts and are able to deal with low nutrient conditions. Their spores were tested concerning e.g. heat-, H2O2- and radiation resistance and the survival under Mars conditions. Our results indicate, that Paenibacillus strains are able to resist extraordinary extreme conditions and that more effort is necessary to fully understand their role in the huge field of planetary protection.

THE INFLUENCE OF STELLAR XUV RADIATION ON PLANETARY ATMOSPHERE

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Planets orbiting at close distances to their host stars are exposed to intense radiation and particle fields. This leads to thermal and non-thermal escape processes of planetary atmospheric material. In close orbits tidal effects become important, because the Roche lobe distance is located closer to the planetary surface and atmospheric particles above this limit are lost to space. The recently discovered super-Earth GJ 1214b is orbiting an M-type star at the very close distance of 0.014 AU. The star is rather inactive and supposed to be several Gyrs old, so the current XUV flux received by the planet is rather low. However, it is well known that young M dwarfs are usually very active, with high XUV fluxes and frequent, powerful flare events. This raises the question on how efficiently thermal escape influenced GJ 1214b during the young phase of the host star. We present energy-limited escape calculations, which take into account Roche lobe effects, heating efficiencies, and the evolution of the stellar XUV luminosity, in order to explore possible scenarios of the evolution of GJ 1214b.
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Pulsar planets are planets that orbits pulsars. Pulsar planets are discovered by pulsar timing measurements through variations in the pulsar spin period. Several planets are already discovered and confirmed around pulsars PSR B1257+12 and PSR B1620-26. Pulsar PSR B0329+54 is still awaiting confirmation of two planets. This paper considers methods of detection long and short periodic planets by the example of PSR B1620-26 and PSR B0329+54.

COSMIC MASERS AS TRACERS AND DIRECT INDICATORS OF STAR FORMATION
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In the Galaxy, microwave radiation can be amplified in the interstellar medium in the immediate neighborhood of young stellar objects, or circumstellar envelopes around evolved stars, resulting in cosmic maser emission. In the star formation regions there are different types of maser sources that trace different youngest evolution stage of protostars. The OH, H2O, methanol masers and others are now recognized to be excellent tracers of star formation. To better understand the nature of star formation activity in the star formation regions of the Galaxy, we have used these classes of masers combined with other astrophysics data to study prominent sites of ongoing star formation in the nebulae.

ROLE OF SPACE DUST ANALOGS IN ABIGENIC SYNTHESIS OF OLYGOPEPTIDES AND NUCLEOTIDES UNDER ACTION OF VUV-IRRADIATION
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All Solar system objects, such as planets, satellites, rings, comets, asteroids, meteorites, and interplanetary dust particles (IDP) are subjected to energetic processing by different kinds of open space energy. Study of the role of charged particles, ultraviolet radiation of different wavelengths, and other energy sources in the abiogenic synthesis of biologically significant compounds (BSC) is closely related to the exobiological investigations in the orbit of Earth related to chemical evolution and the issue of the origin of life on Earth and throughout the Universe. According to recent views, the majority of BSCs may have been introduced to the primeval Earth by micrometeorites, meteorites, and comets. These cosmic objects contain large quantities of complex organic compounds, including carbohydrates, amino acids, and heterocyclic bases of nucleic acids. The problem is the way of their origin. The aim of this work is to study the influence of mineral substrates on the reaction of nucleoside phosphorylation by an inorganic phosphate and oligomerization of amino acids under the action of vacuum ultraviolet (VUV) radiation with wavelengths <200 nm, one of the main energy sources of the Sun. Dry mixture of amino acids (Gly+Phe) and nucleoside with inorganic phosphate (Cyt+iP) were exposed to VUV-irradiation (145 nm) with different doses (up to 2.1x106 J/m2). Gly-Gly, Phe-Gly, Gly-Phe, Phe-Phe, Gly-Gly-Gly, Phe-Gly-Gly, Gly-Gly-Phe and Phe-Phe-Phe were the main products in the case of amino acids mixture with yield up to 0.6%. The presence of mineral components (olivine, piroxene, SiO2) have different influence on the yields and
products of the reaction oligomerization. The abiogenic synthesis of nucleotides from nucleoside and inorganic phosphate also has been observed with summary yields up to 6%. Our experiments indicate that oligopeptides and nucleotides could be synthesized on the surface of small bodies in the Solar system on the early stage of its evolution and safely transported to the Earth.

MEAN-MOTION RESONANCES AND THE PROPERTIES OF PROTOPLANETARY DISCS

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Mean-motion resonances are ubiquitous phenomena in the planetary systems. They carry an important information about the formation processes and the further evolution of those systems. Moreover, their occurrence may increase a chance to detect planets which happened to be in a resonant configuration. In this talk we investigate yet another potential role, which the mean-motion resonances may play in our understanding of planet formation, namely how well we can constrain the physical conditions in the protoplanetary discs using the information about the most common resonant configurations observed in existing planetary systems.

IDENTIFICATION OF MICRO-BIOFOSSILS IN SPACE DUST

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Balloon-borne cryosampling of the stratosphere enables recovery of fragile interplanetary dust particles (IDPs) with their structure and carbonaceous matter largely intact. SEM studies of texture and morphology of particles in the Cardiff collection, together with EDX identifications, shows two main types of putative bio-fossils – firstly organic-walled hollow spheres around 10 microns across, similar to those found in carbonaceous chondrite (CC) meteorites and terrestrial sedimentary rocks and termed ‘acritarchs’. Though acritarchs in both IDPs and CCs probably derive from comets, those in IDPs are less geochimically altered than those in the Orgueil CC and have mineral coatings. Some coverings of unstructured filmy material are seen, suggesting extracellular polymer substances, while some acritarchs have attached fibres. The fibres form a second type of microfossil, being siliceous material but differing from mineral cometary silicates. They resemble siliceous fragments of diatoms, from submicron whiskers up to 10-15 microns long fibres of 1-2 microns diameter, both singly and in complexes. These samples add weight to the earlier proposal that cometary ices are a habitat for diatoms. The discovery of probable micro-biofossils in space dust opens up a new field for astrobiological studies, offering better preserved specimens than in CCs.