Venera-D Landing Sites selection and Cloud Layer Habitability Workshop. October 2-5, 2019. IKI, Moscow, Russia

Observation of the life cycle of extremophilic bacteria of the genus *Acidophillus* under the conditions of the reconstituted Venus atmosphere

Grzegorz P. Słowik

„Bacteria are intelligent little organisms. They can survive almost anywhere and quickly adapt to new conditions”

Satish Nair
Biochemistry professor
University of Illinois

https://phys.org/news/2017-08-dangerous-bacteria-effective.html

UNIVERSITY OF ZIELONA GÓRA

To many questions about Venus that are still unanswered [Glaze et al., 2018] there should also be added an astrobiological question about the possibility of life occurring in the clouds of Venus, as well as its possible form.

In the past, the planet could have been rich in water and had a hospitable climate for life [Way et al., 2016].

It is known that in the lower layer of Venus clouds, from 47.5 to 50.5 km above its surface, there is a pressure of approximately 1 atm, and the temperature is about 60°C [Limaye et al., 2018]. Under such conditions, acydophilic bacteria of the genus Acidithiobacillus (versus Thiobacillus), which are capable of surviving in an acidic environment with a $pH < 3$, are able to survive. Moreover, they are capable of existing in anaerobic conditions [Pronk, 1992; Valdés, 2008]. Anaerobic respiration of Acidithiobacillus ferrooxidans is based on iron (Fe^{3+}). They can also use other electron donors, which are derived from sulfur S^0 and hydrogen H^2 [Ohmura, 2002].

The authors of this study postulate to test the hypothesis of the existence of an atmospheric habitat of acidophilic microorganisms (terrestrial type) in laboratory conditions using the restored and controlled composition and physical parameters of the Venus atmosphere in multifunctional bioreactors.

A crucial research problem is to define the universal chemical „life marker”, which is related to the life activity of potentially living in Venusian clouds - chemoautotrophs. In the case of terrestrial life, its chemical markers, due to the nature of metabolism are: oxygen (O_2) and methane (CH_4). Oxygen consumption prevents an „oxygen disaster” whereas the detected methane level is the effect of both volcanism and the effective metabolism of anaerobic bacteria in higher organisms.

Short description of the research project:

The authors of the study postulate that in the case of Venus, sulfuric acid (H_2SO_4) - the product of chemoautotroph metabolism - can be considered as such a marker. Studies with the strain Acidithiobacillus ferrooxidans, whose UV spectra correlate highly with the spectra recorded for the Venus atmosphere in the same wavelength range λ of electromagnetic radiation, have been started.

The laboratory experimental tests will be used in the research carried out under the conditions close to Venus atmosphere. Their results should at least partialy help to explain the research issues presented above.

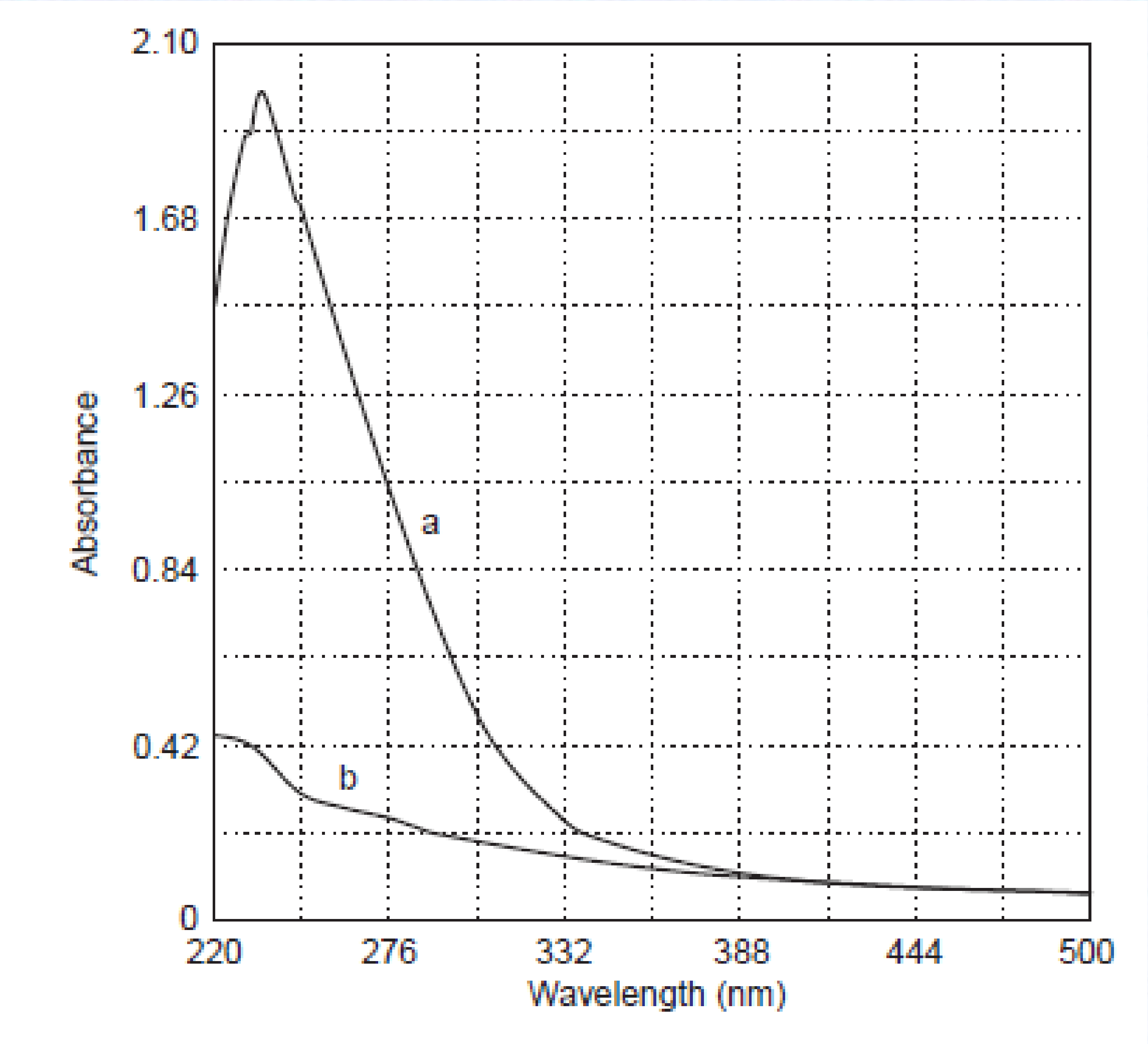

Fig. 1. UV spectrum of iron-sulfur proteins obtained after growth of *T. ferrooxidans* on medium containing (a) iron sulfate heptahydrate, (b) sulfur.

A.B. Więckowski, G. Słowik, J.A. Gąsiorek, P. Gąsiorek, F. Domka, A. Perkowska, *EPR study and structural aspects of ferredoxins obtained from Thiobacillus ferrooxidans*, Applied Microbiology and Biotechnology, 1999, 52, p. 98.

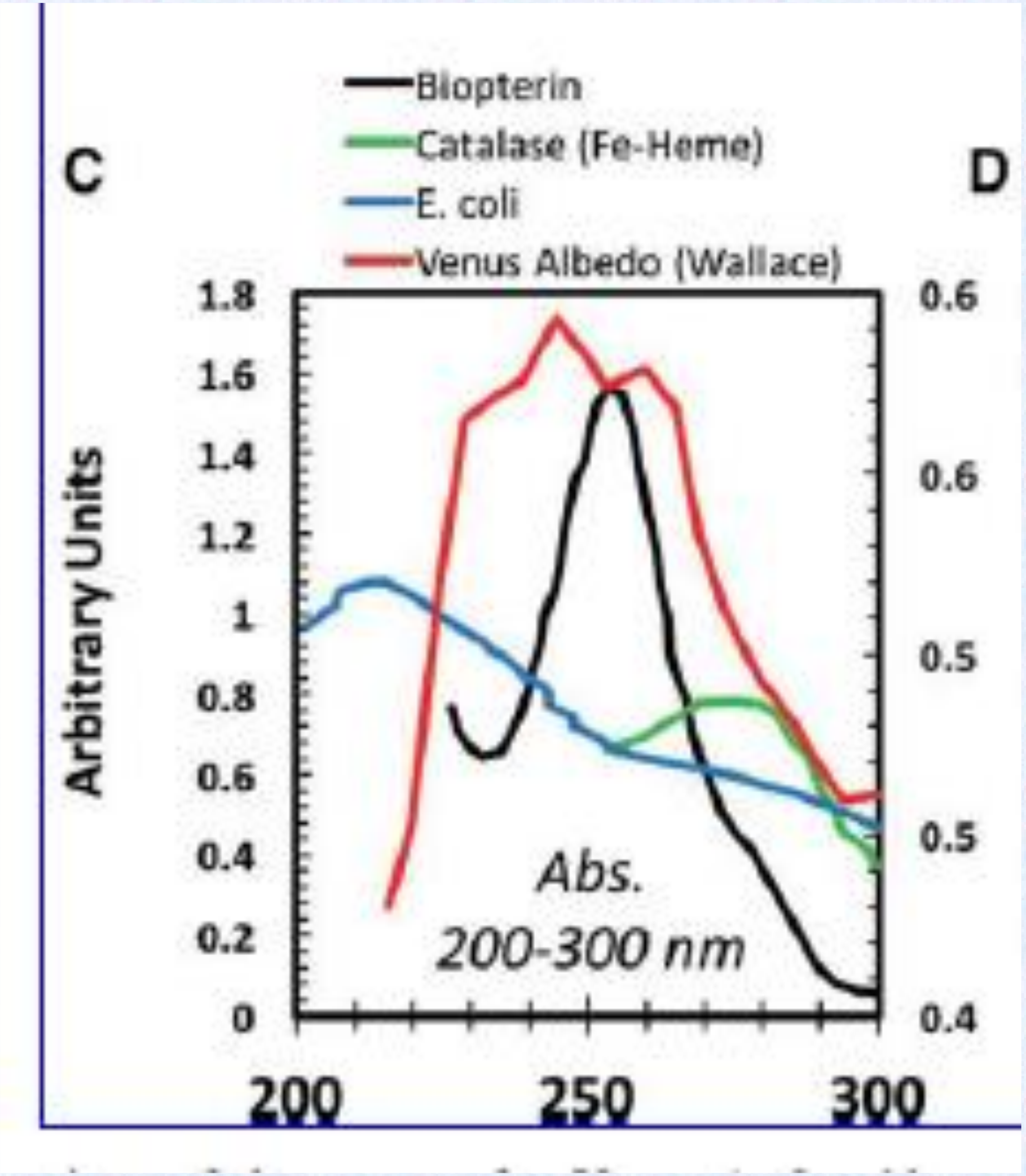

Fig. 2.

S.S. Limaye, R. Mogul, D.J. Smith, A.H. Ansari, G.P. Słowik, and P. Vaishampayan, *Venus' spectral signatures and the potential for life in the clouds*, Astrobiology, 18, 9, 2018, p. 1189.

The bacterium Acidithiobacillus ferrooxidans is very common in nature [Zhang S., 2018] and inhabits various types of (from the geoclimatic side) natural environments characterized by low pH [Quatrini, 2019]. They are found in sulphated soils and rocks, but also in mine waters [Consensus Document, 2006]. The optimal pH for most strains of Acidithiobacillus ferrooxidans is about 2 [Quatrini, 2019]. The minimum pH of the environment in which Acidithiobacillus ferrooxidans increases is in the range of 1.3 to 1.5, depending on the strain of this bacterium [Johnson, 2007]. The intracellular pH of Acidithiobacillus ferrooxidans is close to neutral and is 6.5 [Quatrini, 2019]. These bacteria obtain the carbon necessary for the biosynthesis of cellular material by assimilating carbon dioxide [Campodonico, 2016] from the atmosphere in the Calvin-Benson-Bassham cycle (CBB) [Gale 1967; Esparza, 2010; Quatrini, 2019] and they can bind atmospheric nitrogen [Valdés, 2008]. Both of these substrates of Acidithiobacillus ferrooxidans metabolism occur in the atmosphere of Venus.

Types of Acidithiobacillus ferrooxidans strains

	Strain	Authors, year of publication
1.	2Y	Harneit et al., 2006
2.	A1	Leduc et al., 1997
3.	A2	Harneit et al., 2006
4.	AK1	Braddock et al., 1984
5.	A-4	Harneit et al., 2006
6.	AP19-3	Sugio et al., 2003
7.	ATCC 13661	Valkova-Valchanova et al., 1994; Baldi et al., 1992
8.	ATCC 14859	Kaewkannetra et al, 2009
9.	ATCC 19859	Harneit et al., 2006; Brasseur et al., 2004; Holmes et al., 2001; Barron et al., 1990; Brahmaprakash et al., 1988; Espejo et al., 1987
10.	ATCC 21834	Pronk et al., 1991
11.	ATCC 23270	Ai et al., 2018; Kocaman et al., 2016; Dekker et al. 2016; Liu et al., 2015; Bustamante et al., 2014; Bustamante et al., 2012; Ponce et al., 2012; Thurston et al., 2010; Esparza et al., 2010; Nieto et al., 2009; Harneit et al., 2006; Valdés et al., 2008; Yarzábal et al., 2002a; Giudici-Orticoni et al., 2000; Sampson et al., 1999; Ronk et al., 1991
12.	ATCC 33020	Yarzábal et al., 2003; Yarzábal et al., 2002a; Yarzábal et al., 2002b; Appia-Ayme et al., 1999; Oppon et al., 1998
13.	ATCC 53993	Ramos-Zúñiga et al., 2018; Bustamante et al., 2014; Orellana et al., 2011
14.	B-458-Cu	Kondrat'eva et al., 2002
15.	B-86	Belyi et al., 2006
16.	BRGM	Giudici-Orticoni et al., 2000
17.	BKM B-458	Kondrat'eva et al., 2002
18.	BY-3 (CCTCC-M203071)	Chen et al., 2011; Yan et al., 2010
19.	BYQ-12	Yan et al., 2017
20.	C-52	Harneit et al., 2006; Gehrke et al., 2001
21.	CCM 4253	Kucera et al., 2016; Pakostova et al., 2013ab; Ceskova et al., 2002
22.	CCTCC M20405	Yang et al., 2008
23.	ConTf	Mason et al., 2002
24.	CUMT-1	Feng et al., 2015; Feng et al., 2012a
25.	D2	Leduc et al., 1997
26.	D6	Leduc et al., 1997
27.	D7	Leduc et al., 1997
28.	DECp	Harneit et al., 2006
29.	DLC-5	Xu et al., 2019
30.	DSM 583	Harneit et al., 2006; Sampson et al., 1999; Baillet et al., 1998
31.	DSM 584	Africa et al., 2013
32.	DSM 14882	Govender et al., 2015; Tan et al., 2012
33.	DSM 16786	Latorre et al., 2016
34.	DSMZ 583	Haghshenas et al., 2009
35.	F1	Leduc et al., 1997
36.	FC1	Fowler et al., 1999, Harvey et al., 1997
37.	FY-3	Leng et al., 2009
38.	GF	Yang et al., 2013
39.	H-3	Leng et al., 2009
40.	KCTC 2677	Soo et al., 2002
41.	LR	Bevilaqua et al., 2002; Novo et al., 2000
42.	MAL 4-1	Devasia et al., 2010; Natarajan et al., 1994
43.	MON-1	Sugio et al., 2003
44.	NatTf	Mason et al., 2002
45.	N1	Leduc et al., 1997
46.	N2	Leduc et al., 1997
47.	NCIB 8455	Bacon et al., 1989
48.	PD-2	Lei et al., 2012
49.	PH	Harneit et al., 2006
50.	PTCC 1647	Haghshenas et al., 2009
51.	R1	Harneit et al., 2006; Gehrke et al., 2001; Leduc et al., 1997
52.	R2	Rojas-Chapana et al., 2001
53.	R6	Harneit et al., 2006
54.	R7	Harneit et al., 2006; Gehrke et al., 2001
55.	R10	Harneit et al., 2006
56.	S2	Leduc et al., 1997
57.	SPIII/3	Harneit et al., 2006; Gehrke et al., 2001
58.	SPIII/7	Harneit et al., 2006
59.	SUG2-2	Sugio et al., 2003
60.	T23-3	Kawabe et al., 2003
61.	TFAs2	Kondrat'eva et al., 2002
62.	TFBk	Kondrat'eva et al., 2002; Ageeva et al., 2003; Ageeva et al., 2001
63.	TFBk-Cu	Kondrat'eva et al., 2002
64.	TFI	Kondrat'eva et al., 2002
65.	TFI-Fe	Kondrat'eva et al., 2002
66.	TFN-d	Ageeva et al., 2001; Kondrat'eva et al., 2002; Ageeva et al., 2003
67.	TFD	Kondrat'eva et al., 2002
68.	TFG	Kondrat'eva et al., 2002
69.	TFI-35	Bhatti et al., 1993
70.	TFO	Ageeva et al., 2001; Kondrat'eva et al., 2002; Ageeva et al., 2003
71.	TFL-2	Ageeva et al., 2001; Kondrat'eva et al., 2002; Ageeva et al., 2003
72.	TFNi-3	Kondrat'eva et al., 2002
73.	TFR1	Kondrat'eva et al., 2002
74.	TFV-1	Ageeva et al., 2001; Kondrat'eva et al., 2002; Ageeva et al., 2003
75.	TFV-1-Cu	Kondrat'eva et al., 2002
76.	TFKm	Kondrat'eva et al., 2002
77.	TFN	Kondrat'eva et al., 2002
78.	TFP	Kondrat'eva et al., 2002
79.	TFT	Kondrat'eva et al., 2002
80.	TFUch	Kondrat'eva et al., 2002
81.	TFUd 2	Kondrat'eva et al., 2002
82.	TFUd 3	Kondrat'eva et al., 2002
83.	TfW	Das et al., 1998; Das et al., 1997
84.	TfCu20	Das et al., 1998
85.	TFWc	Kondrat'eva et al., 2002
86.	TKY-2	Leng et al., 2009
87.	TFZ	Kondrat'eva et al., 2002
88.	TM	Silverman et al., 1959

The Acidithiobacillus ferrooxidans bacterium can also grow by being powered by electrons from the cathode and anode electrodes, according to the reactions [Yamanaka, 2008]:

$$2H^+ + 2e \rightarrow H_2 \quad \text{(with cathode)} \qquad (1)$$

$$2Fe^{2+} - 2e \rightarrow Fe^{3+} \quad \text{(with anode)} \qquad (2)$$

The process of oxidation (bioxidation) of ferrous ion and pyrite by the bacteria Acidithiobacillus ferrooxidans, through which they gain the energy released in this way, are represented by chemical reactions (3) and (4) [Yamanaka, 2008]:

a) (bio) oxidation of ferrous ion

$$2Fe^{2+} + 2H^+ + 0.5O_2 \rightarrow 2Fe^{3+} + H_2O \qquad (3)$$

b) (bio) oxidation of pyrite

$$FeS_2 + 3.5O_2 + H_2O \rightarrow Fe^{2+} + 2SO_4^{2-} + 2H^+ \qquad (4)$$

Acidithiobacillus ferrooxidans can remove hydrogen sulfide (H_2S) from the environment [Halfmeier, 1993, Part 1] in a two-step process presented by reactions (5) and (6):

$$H_2S + Fe_2(SO_4)_3 \rightarrow S^0 + 2FeSO_4 + H_2SO_4 \quad (5)$$

$$4FeSO_4 + O_2 + 2H_2SO_4 \rightarrow 2Fe_2(SO_4)_3 + 2H_2O \quad (6)$$

Fig. 3. Measuring facilities set.

Fig 4. Liquid bacterial cell culture in bioreactors.

Matrix of the Life (ML)

Acidithiobacillus ferrooxidans, Venus

$$\begin{pmatrix} Earth - like\ atmospheric\ pressure & presence\ in\ the\ atmosphere:\ CO2, N2, Fe, S & the\ occurrence\ from\ 47.5\ to\ 50.5\ km\ above\ the\ surface \\ temperature\ on\ Earth: 60\ °C & presence\ in\ the\ atmosphere\ of\ condensation\ centres & the\ occurrene\ of\ free\ current\ carriers\ in\ the\ atmosphere \\ pH < 3 & Gas\ circulation\ in\ the\ atmosphere & occurrence\ of\ such\ phenomena\ as\ volcanism\ and\ sand\ storms \end{pmatrix}$$

Elements of Matrix of the Life

a11 - the Earth-like atmospheric pressure

a21 – temperature around 60° C

a31 – pH < 3

a12 - presence of $CO2, N2, Fe, S$ in the atmosphere

a22 – presence of condensation centres in the atmosphere

a32 – gas circulation in the atmosphere

a13 – occurrence from 47.5 to 50.5 km above the surface

a23 – occurrence of free current carriers in the atmosphere

a33 – occurrence of such phenomena as volcanism and sand storms

Thank you for attention!