1. PUBLISHABLE SUMMARY

The DECAVE project was a two-year EU FP7 individual fellowship project running from 2014 to 2016. Its primary goal was a detailed geochemical, mineralogical, geomicrobiological, morphological and microbiological characterization of secondary mineral deposits, or speleothems, found in caves for understanding their nature and origin. This involved an approach that was simultaneously integrative, multidisciplinary and multi-scaled. Until recently many of the cave studies solely relied either on molecular biology analyses or morphological and mineralogical characterization. Consequently, more complete and comprehensive multidisciplinary studies on the geomicrobiology and biogeochemistry of caves, and biosignature identification are needed, and the DECAVE project addressed this need. The interest of this project's concept lies in the fact that caves on Earth may represent outstanding examples of ongoing geological, ecological and biological processes, where we can search for: (i) biosignatures, or traces of life, in the rock record for understanding the origin of life on Earth and possibly on other planets, (ii) paleoenvironmental changes, (iii) biomineralization activities; (iv) subsurface microbial diversity, (v) new bacteria and metabolic pathways, (vi) antimicrobial secondary metabolites, (vii) the role of microorganisms in biogeochemical cycles, and (viii) the preservation of prehistoric rock art.
The concepts and approach developed in this project involved a series of research objectives comprising four work packages:
WP1. Morphological, geochemical and mineralogical characterization of secondary mineral deposits from subsurface rock environments, including lava tubes from Canary Islands (Spain), Easter Island (Chile) and Galapagos Islands (Ecuador).
WP2. Identification of microbial communities associated with mineral deposits using powerful new molecular tools.
WP3. Appraisal of microbe-mineral interactions to assess biogenicity and elucidate on the role of microorganisms to mediate mineral precipitation.
WP4. Recognition of biominerals, microbial fabrics or fossil microorganisms as biosignatures valuable for astrobiology.

By establishing national and international collaborations with different world-leading research groups, it was possible to perform an inter- and multi-disciplinary approach, essential for obtaining significant results and achieving the main goals of the DECAVE project.

A multiproxy approach based in the combination of microscopy, mineralogy, analytical pyrolysis (Py-GC/MS), pyrolysis compound specific isotopic analysis (Py-CSIA) and light stable isotope analyses showed that coralloid stalactites from the Ana Heva lava tube in Easter Island consist of three major coloured layers with different mineralogical composition. Their genesis was related to two different stages of speleothem development caused by climate changes on Easter Island. Different water regimes and an increase on the average temperature during speleothem growth were recorded by Py-CSIA and isotope analyses, corroborating the so-called climate warming during the Holocene.

Among the important findings are the microbial diversity associated with secondary mineral deposits from Canary and Galapagos Islands. The 16S rRNA gene analyses showed that Actinobacteria and Proteobacteria dominated in these lava tube cave systems. Most of the phylotypes associated with

anamiller@irnas.csic.es

AVDA. REINA MERCEDES,10
41012 SEVILLA
TEL: +34 95 4624711
FAX: +34 95 4624002

these lava tube speleothems were affiliated to chemoautotrophs, such as *Ferrithrix thermotolerans*, and other mineral utilizing microorganisms like *Aciditerrimonas ferrireducens*, *Desulfuromonas* sp. and *Desulfovibrio* sp., suggesting that many of the phylotypes identified are adapted to extreme subsurface environments. Moreover, many of the clone sequences were unassigned to classified species when using SILVA database, indicating that these subsurface environments represent a potential source of novel taxa and metabolic pathways.

The suitability of cave speleothems as biosignature repositories was investigated for samples collected in Canary, Easter and Galapagos Islands. Microbial mats, extracellular polymeric substances and mineral deposits bearing Si, Ca or Fe, tubular empty sheaths, mineralized cells, filamentous fabrics, as well as "cell-sized" etch pits or microborings produced by bacterial cells were commonly observed. These features evidence microbe-mineral interactions and represent mineralogical and microbial signatures of life. Hence, we propose lava tubes of Canary, Easter and Galapagos Islands as suitable environments to acquire valuable data for comparative astrobiological studies between Earth's caves and Mars subterranean environments.

In summary, the results obtained within the DECAVE project allowed a deeper understanding on the nature and origin of secondary mineral deposits from lava tubes of Canary Islands (Spain), Easter Island (Chile) and Galapagos Islands (Ecuador), as well as the recognition of biosignatures valuable for astrobiology. In addition to novel insights in cave deposits, this individual research project contributed to better understand the adaptability of extremophiles to hostile rock environments and their interaction with minerals. We expect that the dissemination of the DECAVE project results contribute for further studies on the search for traces of life in the geological record of the Earth's subsurface and possibly on other planets, as well as to estimate the plausible habitability of Mars subsurface.

The dissemination and transfer of knowledge activities during the reporting period (24 months) comprised 7 papers in international journals included in SCI, 1 article under revision, 1 submitted and 2 in preparation. In addition, 3 peer-reviewed book chapters have been published. The fellow has participated in 22 national and international conferences, 1 as invited speaker (*22nd International Karstological School,* Slovenia) and 6 will be presented after the execution of the DECAVE project.

Moreover, the development of the DECAVE project has provided:

- Remarkable mastery in microbiology and molecular biology techniques, scanning electron microscopy, and analytical techniques for geochemical and mineralogical characterization.
- Co-operation through joint experimental activities with national and international researcher groups involving further co-authorship of journal articles.
- Coordination, supervising and leadership skills through the supervision of 2 PhD students, 1 PhD student research stay at the host group, coordination tasks in the laboratory, and DECAVE project management.
- Transferable skill training and reinforcement of communication, initiative and social skills by disseminating the relevant results of the DECAVE project in international scientific meetings and seminars at the host Institute and international institutions during short-term research visits.
- Significant scientific maturity and expertise essential to be awarded with a 2-year postdoctoral contract in the framework of the *Juan de la Cierva* programme of the Spanish Ministry of Economy and Competitiveness (MINECO). This contract has recently started and will allow her to continue a promising scientific career at the host institution.

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anamiller@irnas.csic.es

AVDA. REINA MERCEDES,10
41012 SEVILLA
TEL: +34 95 4624711
FAX: +34 95 4624002