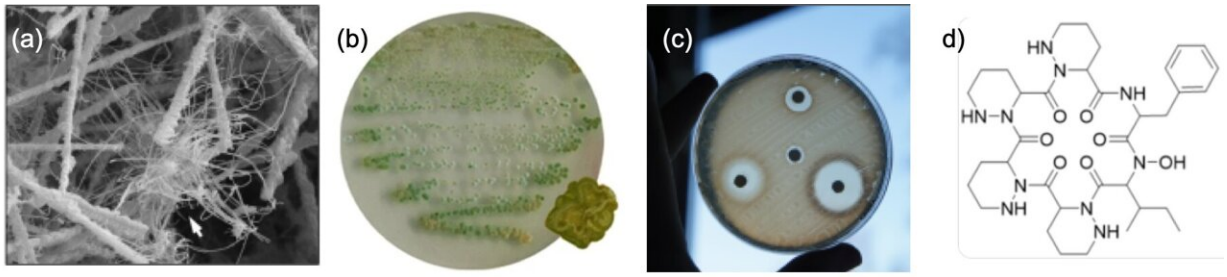
Lunaemycin, a new antibiotic extracted from moonmilk deposits

February 2 2023



The different stages of the search for bioactive molecules produced by moonmilk bacteria. (a) Visualization of filamentous bacteria between moonmilk fibers. (b) Bacteria isolated from moonmilk (Streptomyces lunaelactis). (c) Antibiotic activity of Streptomyces lunaelactis. (d) Structure of lunaemycin. Credit: University of Liège / S.Rigali

A study conducted by scientists from the University of Liège and the HEDERA-22 spin-off on moon milk—a mineral deposit found in caves and used for its curative properties—has led to the discovery of a cryptic compound active against bacteria that are multi-resistant to antibiotics. This discovery is the subject of a technology transfer and a publication in the *International Journal of Molecular Sciences*.

Since the dawn of time, human beings have been able to draw on the environment for resources, techniques and sometimes beliefs to protect themselves from disease and to care for their health. While the

effectiveness of certain ancestral practices has been demonstrated and endorsed by modern medicine, the rationality of many traditional medicines remains misunderstood and even suspect.

This is the case with the use of moonmilk, a concretion frequently found in various forms (pasty, dry or liquid) in limestone caves, a remedy used for curative purposes. "This speleothem is most often observed in the form of a soft rock depending on its hygrometry, a sort of mineral cheesecake," explains Sébastien Rigali, a molecular microbiologist at the Centre for Protein Engineering—CIP (InBios/Faculty of Science) of the ULiège. "There is a lot of archaeological evidence of its use as an anti-infectious agent, mainly in the Swiss and Austrian Alps."

The laboratory of Dr. Sébastien Rigali, F.R.S-FNRS Research Director, decided to study the microbial flora of moonmilk out of simple curiosity, in order to find a rationale for its use in human and animal therapy.

"The first step was to go into the field, into the caves of the Condruzian plateau, in order to find deposits of moonmilk and to isolate filamentous actinobacteria, the bacteria that are champions in the production of antimicrobial agents. The researchers found many of them, both in number and diversity. All the most prolific bacterial genera in terms of antimicrobial production are represented."

Analysis of the genomes of the isolated bacteria revealed that they participate together in the constitution of a vast subterranean pharmacy. This microbial consortium is capable of producing hundreds of antibiotics, some of which are used daily today. But what is probably the most fascinating is that the vast majority of the biosynthesis genes involved in bioactive compound production found in these bacteria are cryptic, i.e. it is not possible to associate a known molecule with them.

Expeditions to search for and collect moonmilk deposits in the Walloon Karst. Credit: Vincent Gerber

This means that these strains constitute a real reservoir for the discovery of new bioactive molecules. "Statistically, it's as if all you have to do is bend over and pick up and identify a new antibiotic, antifungal or even anticancer agents," says the researcher.

The reality is obviously much more complex. These molecules do not lend themselves easily to being grown under laboratory conditions. Extracted from extreme development conditions and oligotrophic, it is difficult to keep them 'alive' once they are brought to the surface. Moreover, their development in cultivation media is often not optimal, the media generally used being too rich, and therefore to a certain extent toxic, compared to the nutritive condition found in their original

ecological niche.

It is through collaboration between the laboratories of the CIP, the GIGA, the Mass Spectrometry Laboratory and the CIRM of the University of Liège and the company HEDERA-22, a ULiège spin-off active in the discovery and production of biomolecules of therapeutic or agro-industrial interest, that these efforts have paid off and have made it possible to reveal a first cryptic compound.

"It is an antibiotic named 'lunaemycin' and produced by a new bacterium Streptomyces lunaelactis, lunaemycin and lunaelactis referring to the ecological niche from which this molecule and this bacterium originate, the moonmilk. The lunaemycin has interesting properties, particularly active against Gram-positive bacteria that are multi-resistant to antibiotics."

Other criteria still need to be evaluated to know whether this new antibiotic will one day be exploitable, but lunaemycin represents above all a first shot in the submerged part of the 'moonmilk iceberg', which contains many molecules that are still unknown to date and which could have similar properties. The lunaemycin is also this first 'proof of concept' that demonstrates the capacity of research teams to discover new molecules, from the first contortions in the galleries of the Walloon caves to their structural resolution and the determination of their biological activities.

All the skills of Sébastien Rigali's laboratory and the laboratories of the University of Liège involved in this research have been transferred to the spin-off HEDERA-22, whose objective is to exploit the potential of the bacteria isolated from moonmilk and to reveal the most promising molecules in human therapy but also in the agro-industrial field. If this study brings a certain legitimacy to the use of moonmilk in traditional medicine, it demonstrates once again the extent of the treasures that the

microbial world is still ready to offer us.

More information: Loïc Martinet et al, Lunaemycins, New Cyclic Hexapeptide Antibiotics from the Cave Moonmilk-Dweller Streptomyces lunaelactis MM109T, *International Journal of Molecular Sciences* (2023). DOI: 10.3390/ijms24021114

Provided by University de Liege