

Tardigrades: A gateway to understanding extremophile resilience

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Abstract

Tardigrades, also known as water bears, are microscopic organisms that exhibit an extraordinary ability to survive extreme environmental conditions. In this work, we studied the mechanisms permitting their extreme temperature, desiccation, radiation, and high-pressure survival through experimental assays and molecular analysis. Here, we demonstrate that tardigrades employ a unique combination of cryptobiosis, protective protein synthesis, and efficient DNA repair systems to survive lethal conditions, enabling them to survive unmatched environmental challenges unlike any other organism. Thus, we emphasize the evolutionary implications with their themes, to shed light on understanding life not only on Earth but also elsewhere. This work contributes to our ever-expanding understanding of extremophiles and deconstructs crucial processes of resistance and adaptation within life forms.

Keywords: Tardigrades, extremophile resilience, water bears

Introduction

Tardigrades, scientifically classified under the phylum Tardigrada, are often known by the common name water bears. An example of species in this group is *Milnesium tardigradum*. Johann August Ephraim Goeze, a German zoologist, was the first to report seeing the animals in 1773 as "little water bears." Three years later, the Italian biologist

Lazzaro Spallanzani would give them the official name of Tardigrada, the name that described their characteristic movement-slow walker. These tiny invertebrates belong to the group Articulata, which spread across most parts of the ecosystems on earth and their habitats spread from the deepest oceans to the summits of the highest mountains.^[1]



Source: 40 Tardigrade Facts About the World's Toughest Animal - Facts.net

The microscopic size of the bodies goes up to 2.1 mm, normally in a cylindrical body shape with five distinct portions: the head and four others, each of which has two clawed legs. Bodies are covered with an elastic cuticle which can be smooth or covered with scales or plates. The digestive system is fully developed to accommodate the changing diet due to environment: they eat algae, bacteria, and fungal cells, but others feed on other tiny invertebrates like rotifers and nematodes, or even smaller tardigrades. Their nervous system is advanced with brain-like characteristic around the mouth and with segmented ganglia in the ventral nerve cord along the abdomen. More sophisticated sensory structures like papillae, chemoreceptors, and even eyes complete their features.^[2] The tardigrades generally exhibit sexual dimorphism; most species are even capable of parthenogenesis-an asexual

mode of reproduction in which females produce offspring without mating. Fertilization may either be internal or external where the eggs are protected by a hard shell. Actually, scientists believe that there are approximately 1,200 species of tardigrades; however, the number is not yet confirmed. A general division of this organism is into two: Eutardigrada, which contains freshwater and terrestrial species, while Heterotardigrada is composed of marine and terrestrial species.

The life cycle of these organisms involves hibernation as they acquire a particular set of characteristics when they are inside a dormant state. Hibernation means becoming dormant, sleeping, resting, and the organisms have short body structure, generally between 0.1 to 5mm in length.

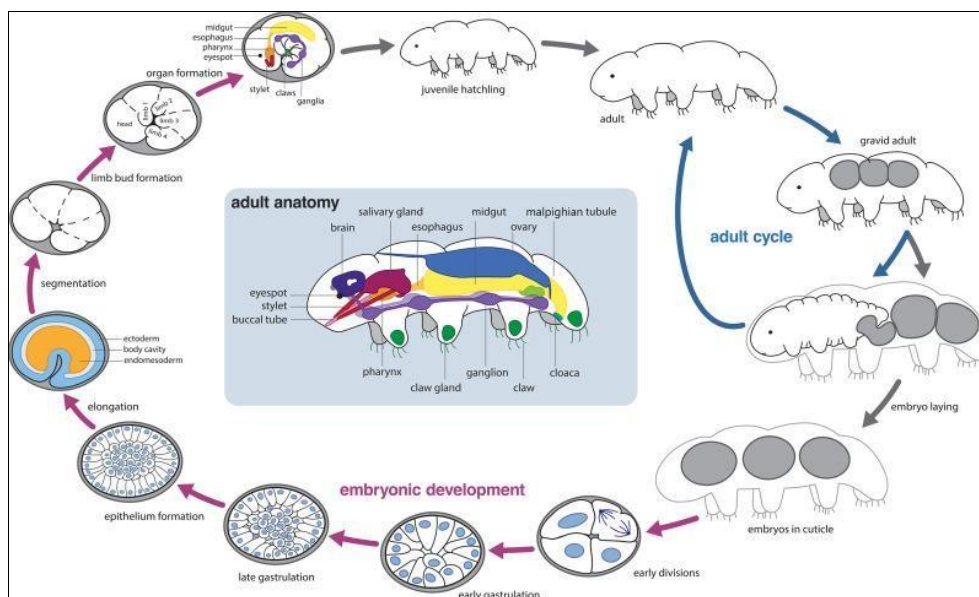
There are three stages of life for tardigrades: egg, juvenile, and adult.

- 1. Egg Stage:** Tardigrades are egg laying species and typically encapsulate the eggs in their shed cuticle. The number of eggs varies from a few tens to several dozens for the species. These eggs possess a hard shell, protecting them against stressful conditions. The incubation period depends on the species and environmental conditions and can last from several days up to weeks, during which the embryo develops its basic anatomical features.
- 2. Juvenile Stage:** The juvenile tardigrade comes out as if it were a miniature adult, a process known as direct development. Tardigrades are unlike any other animal in the short period they live, as they experience very little transformation from being born; they look almost exactly the same before birth as after. They grow by means of ecdysis through an insect-like process—a type of

exuviation, where they repeatedly shed their outer cuticle and can even molt up to twelve times over its lifetime, growing bigger without adding extra segments to the body.

- 3. Adult Stage:** It is at the adult stage, after undergoing several molts that the tardigrades will be mature and sexually ready, a process of weeks or several months depending on species and environmental conditions including temperature and the presence of food. Adult tardigrades can reproduce either through sexual means in which males fertilize eggs inside the body or externally, or parthenogenesis.

Such good adaptability and especially unique life cycle make tardigrades a subject of big scientific interest. Their role is furthering knowledge about extremophiles—organisms living in some of the most inhospitable places on Earth. [3]



Source: Tardigrades and their emergence as model organisms – ScienceDirect

How are they nearly indestructible?

A phylum, Tardigrada, is a group of diverse species that exhibits rather extraordinary morphological and physiological traits, particularly those that ensure survival in extreme dry and cold conditions. Among nematodes, rotifers, and tardigrades, this is the only group capable of withstanding desiccation and freezing at all stages of its life cycle: egg, juvenile, and adult. Such resistance allows populations of tardigrades to be among the last survivors in some of the most challenging ecosystems and microhabitats. [2]

One of the most impressive adaptations of tardigrades is cryptobiosis - a reversible ametabolic state caused by such factors as desiccation, freezing, low oxygen levels, or high salinity. Cryptobiosis occurs especially often among limno-terrestrial species. Tardigrades have proved to be ideal subjects for research in astrobiology because of their remarkable capacity to survive under various conditions of environmental stress. They have been proven to survive extreme environmental conditions; they can tolerate highly ionizing radiation, temperature fluctuations, and even the vacuum of space. In the tun state, the conditions may enable the tardigrade to live between -328°F (-200°C) and over

300°F (149°C), resistance exposure from radiation, as well as pressures greater than the deepest oceanic trenches. [2] Miraculously, anhydrous tardigrades have been recovered still viable after a ten-day flight in low Earth orbit and returned unscathed following solar ultraviolet radiation and vacuum. Experiments also suggest that desiccated tardigrades can survive hypervelocity impacts traveling almost 3,000 feet per second or 900 meters per second at extreme pressure. [1]

Tardigrades expel over 95% of their body water and withdraw into a dormant state with their heads and legs retracted inside to enter cryptobiosis. The induction process is often triggered by dehydration, freezing, lack of oxygen, and excessive salt. In this state, tardigrades become desiccated spheres whose metabolic activities are reduced to a large extent, and on improved conditions, they rehydrate and reactivate. Indeed, as indicated by studies, entrance and exit from cryptobiosis can involve the production of protective biomolecules containing such specific carbohydrates, proteins, antioxidant enzymes, and free radical scavengers. [1]

An anhydrobiosis state confers resistance to the dehydration of tardigrades but increases their susceptibility to other

physical stressors, such as extreme heat, radiation and various chemicals like ethanol and hydrogen sulfide. Not all species of tardigrades, however, have the same level of desiccation resistance. Within the same species, populations may have different levels of resistance to desiccation. [1]

Anhydrobiosis begins with an induction phase where the tardigrade undergoes metabolic and anatomical changes required for survival in adverse conditions. Such adaptations include body shrinkage in order to minimize surface area and enable the formation of the tun state, hence minimizing evaporation to an approximate 50% loss of liquid water and internal protection in case of desiccation. Only after free water is completely lost do tardigrades begin a procedure for

replacing bound water with bioprotectants, such as trehalose, which stabilizes fundamental macromolecules, such as nucleic acids and proteins, and hence their structure. [1]

Besides trehalose, other protective molecules, including LEA (late embryogenesis abundant proteins), HSP (heat shock proteins), CAHS (cytoplasmic abundant heat-soluble proteins), SAHS (secretory abundant heat-soluble proteins), and aquaporin proteins, are also involved in the process of anhydrobiosis. Such adaptations make a good testimony to the extraordinary robustness of tardigrades and their ability for survival in extreme conditions. [1]

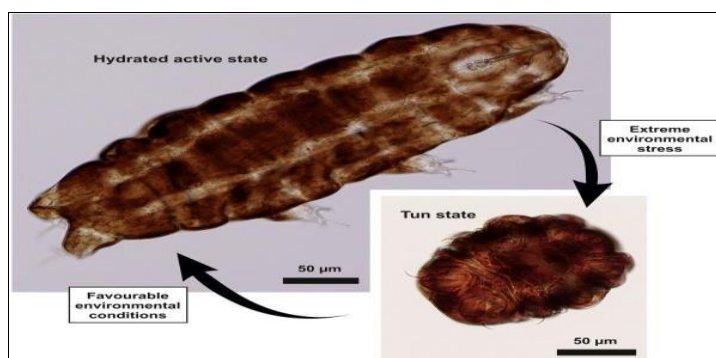


Source: Tardigrades and their emergence as model organisms – ScienceDirect

What do we learn from them and how can we utilize this new information?

The Cell Science-04 experiment of its kind seeks to understand the genetic mechanisms that make it possible for tardigrades, colloquially known as water bears, to thrive in stresses found on Earth as well as during space missions experienced by astronauts. NASA calls such research one step closer to developing protective gear for humankind going on long-duration space missions. The tardigrades sent for this experiment were launched to the International Space Station on June 5, 2021 aboard a SpaceX Dragon cargo spacecraft. Already in 2007, a research team from Europe

had inserted 3,000 living tardigrades into Earth orbit on the exterior of the FOTON-M3 rocket, remaining in orbit around Earth for 12 days where they survived with 68%. [4] Unlike all other experiments that were carried out so far, Cell Science-04 tardigrades will live inside dedicated scientific equipment designed for long-term exposure. Such a setup will allow the remote real-time monitoring and control of the living conditions of the tardigrades, thus providing an extraordinary opportunity to study cellular, tissue, and microscopic animal cultures under space environment conditions in detail. [4]



New insights into survival strategies of tardigrades - ScienceDirect

For one, it is known that water exists in Martian soil (Mitrofanov et al. 2014) while at the Martian pole, the temperature can reach as high as 20 °C during summer (NASA official webpage). The environment in Mars mayn't be the most hospitable for life, but it is known that these tardigrades have a phenomenal survivability both as an anhydrobiotic organism and when it is active. The ability to enter cryptobiosis is essential for their survival, ensuring they can survive a long trip in space and endure long periods of unfavorable conditions. [5]

Investigations on tardigrades may open up the possibility of exploring their potential survival prospects on other celestial bodies in our own solar system, such as planets with moons. However, it is of paramount importance to further investigate the resistance of tardigrades towards combined stressors such as cosmic radiation acting in microgravity, or low temperatures combined with harmful chemicals. Such investigations shall be highly informative for the limits of survival for multicellular organisms from Earth and especially in the search for extraterrestrial life. [2]



Conclusion

Tardigrades, with their extraordinary resilience and adaptability, continue to captivate the scientific community as model organisms for studying extremophiles. Their ability to survive in a wide array of extreme conditions—from desiccation and freezing to high levels of radiation—offers unique insights into biological tolerance mechanisms. This research on tardigrades not only broadens our understanding of evolutionary adaptation but also holds potential applications in fields like astrobiology, medicine, and biotechnology. By unraveling the mechanisms behind tardigrades' stress tolerance, scientists may uncover strategies to protect other organisms, including humans, from harsh environmental stressors. Future research should aim to further decode the genetic and molecular basis of these resilience traits, which may pave the way for innovative advancements in developing life-support systems and preserving biological materials under extreme conditions. The journey into the biology of tardigrades highlights the promise of these microscopic organisms in expanding our knowledge of life's capacity to endure and thrive against formidable challenges.

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