

Anhydrobiosis as an anti-aging strategy in tardigrades: testing the "Sleeping Beauty" hypothesis

The main goal of the project is to investigate the effect of anhydrobiosis on aging of anhydrobiotic organisms, using tardigrades as a model. In this way the Sleeping Beauty hypothesis will be tested. The hypothesis assumes that organisms do not age during anhydrobiosis. Simultaneously it will be possible to check whether anhydrobiosis in tardigrade life cycle is just a passive response to extremal environmental conditions or has a some kind "pro-health" effect, i.e. after anhydrobiosis episode different kinds of damages accumulating during anhydrobiosis, as well as, periods of full activity are repaired.

Water bears (Tardigrada) are a group of small aquatic or/and terrestrial invertebrates inhabiting almost all Earth ecosystems. Tardigrades have typical and well-developed internal organs, i.e. the excretory, reproductive, digestive and nervous systems, while there are no circulatory and respiratory systems. However, the feature that distinguishes tardigrades from most of invertebrates is their ability to undergo cryptobiosis (including anhydrobiosis) at any stage of their life. Due to this ability, tardigrades which are able to survive in extreme and hostile habitats, became a model organisms not only in biology, but also in astrobiology, and even in medicine.

Cryptobiosis is a reversible state in which cell metabolism is extremely reduced. This phenomenon occurs in some invertebrates (including tardigrades) under extreme environmental conditions. The most known form of cryptobiosis is anhydrobiosis defined as ability to survive almost complete loss of body water. In this dehydrated state, tardigrades can survive extremely low and high temperatures, high doses of radiation, extremely high and low pressure or even conditions of a cosmic space. As mentioned above, anhydrobiosis is also supposed to affect the aging process that is illustrated by the Sleeping Beauty hypothesis. So far, research concerning the hypothesis verification has been undertaken occasionally. The same applies to the hypothesis underlying mechanisms. Furthermore, the anhydrobiotic abilities of particular tardigrade species are also poorly known and understood.

We are going to study two tardigrade species differing in anhydrobiosis capability and cultured under the laboratory conditions on Petri dishes, in a specially prepared medium, and in professional environmental chamber. Tardigrades will be fed with rotifers. Experiments concerning tardigrade lifespan and their fertility will be performed on multi-well plates, and tardigrades will be cultured individually, in pairs or in small groups. Petri dishes lined with filter paper imitating the natural conditions prevailing in drying mosses will be used to introduce tardigrades into anhydrobiosis. After determination of basic features of the life history of the studied species (including growth rate, lifespan and fertility), tardigrades will be introduced into different number of anhydrobiosis episodes of different duration to investigate whether anhydrobiosis affects their life history and, above all, their overall lifespan. Using a transmission electron microscope the level of cellular organelle degeneration (regarded as a cell aging marker) during active life and after anhydrobiosis will be determined. In addition, the degree of DNA damage and the level of expression of selected proteins participating in cellular processes considered crucial for aging and/or anhydrobiosis (DNA repair, telomere dynamics, chromatin structure, mitochondrial function and endoplasmic reticulum stress) will be tested. Such comprehensive studies performed at organismal and ultrastructural levels, and concerning anhydrobiosis and its effect on tardigrade life have not been conducted so far. The obtained results will allow therefore to explain the role of anhydrobiosis in tardigrade life history. Confirmation of applicability of the Sleeping Beauty hypothesis in the case of tardigrades will allow for explanation whether these invertebrates can actually avoid the negative consequences of aging. This will open a wide range of possible research on slowing down or even arrest of aging processes in other organisms, including humans. These studies are therefore of great importance not only for understanding the biology of tardigrades, but also for a better understanding of aging processes.

Summarizing, the proposed project will allow to study and better understanding the process of anhydrobiosis in tardigrades and its influence on the life of these small invertebrates. In addition, it will give us a better look at the aging processes of anhydrobiotic organisms, which can be of great importance for our understanding of aging processes in general.