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Biology, Adaptability, and The Economic Applications of Tardigrades In Future

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Abstract: *Tardigrades are small microscopic creatures also known as moss piglets or water bears. They are extremophiles and well known for its survivability. After successfully ruling the space tardigrades are now expected to save lives. From being a 'survivor' tardigrade is now headed to be a 'savior'. This survivability is due to a special type of sugar known as "Trehalose". Trehalose can be found in extremophiles organisms including tardigrades. The unique feature of this sugar is the ability to preserve biological molecules. One of the big applications of the tardigrades are the "dry vaccine". Our world is struggling through a big crisis of covid-19 vaccine, it is next to impossible to make the highest demanded vaccine available to every corner of the earth at the low-temperature range in such a short period of time, and according to WHO half of the vaccines get wasted due to the cold chain method So, we can implement these dry vaccines for covid-19, to reduce the freezing cost, increasing the shelf life of vaccine and make every vaccine reach to needy in a live condition. Now, trehalose is not only confined to preserve vaccines but this can help in preserve the organs that are going to be used either for transplantology or organ donation. This special protein is yet to give a new turn to not only the medical field and to save human life but tardigrades can be implemented for plants in increasing the tolerance to a stressful environment for future climate changes and space settlement hence this paper provides an overview regarding the application and economical aspects of the tardigrades*

Keywords*: Tardigrades,Economic,Importance, Trehalose, COVID-19*

I. Introduction

The tardigrades are small microscopic eight-legged metazoans having four pairs of stubby legs with big claws, an oval stout body with a round back and lumbering gait. They are and also known as moss piglets or water bears. These were discovered first by a German zoologist named Johann August Ephraim Goeze in 1773. Tardigrada ("slow stepper") was the name given later by the Italian biologist Lazzaro Spallanzani. The most mysterious part of Tardigrades is their ability to go into anabiosis. When this process occurs, the tardigrade fold itself like a ball, loses all its water content, and forms a double-walled thick cuticle. Their metabolism processes drop to a very low level.

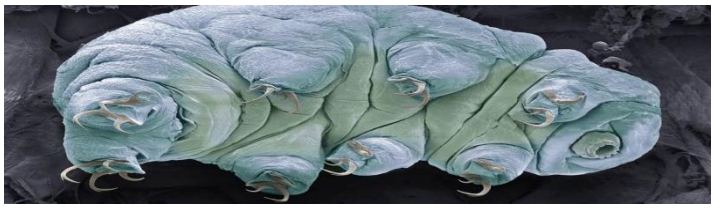

Figure 1 illustrates the lateral view of a tardigrade (Credit: ncbnews.com)

Tuns have been known to survive very harsh environmental conditions such as immersion in helium at -272° C (-458° F) or heating temperatures at 149° C (300° F), exposure to very high ionizing radiation and toxic chemical substances and long durations without oxygen. They are well known to survive in space. It's like they are born with a natural space suite. Without any protection. Some species are identified to survive in complete vacuum and cosmic radiations. The Tardigrades resume their metabolism state within a few hours when the condition is favorable.

The usual habitats are lichens and mosses. They are often detected when a piece of moss is soaked in water. One can find tardigrade in environments such as soil, beaches, sand dunes, and freshwater or marine sediments, (up to 25,000 animals per liter). They are known to live worldwide in moist habitats, along rocky shorelines, and in the bottoms of streams, lakes, Some live-in hot springs and a few live-in symbiotic relationships with or on the bodies of other organisms. Tardigrades use a needle-like mouth to pierce the walls of plant cells and feed on the liquid inside them. Most of the species are plant eaters, but some are predators that feed on tiny invertebrates or bacteria and a few are detritivores, feeding on dead tissue and debris.

Tardigrades are popular due to its unique property of survival. Over 1200 species of the tardigrades have been yet discovered. They are extremophiles and can survive even in lethal conditions. One can find tardigrades from the top of the mountain to the deep sea and from tropical regions to Antarctica. Most species live in the freshwater or semiaquatic terrestrial environment, while about 150 marine species have been recorded. All tardigrades are aquatic habitat because they need water around their bodies to permit gas exchange as well as to prevent desiccation. They are mostly found living in a film of water on lichens and mosses, in sand dunes, soil, sediments, and leaf litter [0]. Tardigrades belong to an elite category of animals known as extremophiles, or critters that can survive environments that most others can't. For instance, tardigrades can live to 30 years without food or water. They can also survive, at zero temperature and the temperature above boiling point, at pressures six times that of the ocean's deepest temperatures as cold as absolute trenches and in the vacuum of space. They contain a unique protein in their bodies called Dsup—short for "damage suppressor"—that protects their DNA from being harmed by things like ionizing radiation, which is present in soil, water, and vegetation.

Anoxybiosis and encasement, are some of the responses one might observe in a variety of organisms. Cryptobiosis is a state where metabolism is stop for a while—an act diagnostic of death. Cryobiotic occurs due to freezing, and anhydrobiosis due to drying. In the latter, the organism surrenders its water content in order to become a desiccated pellet. Both results in the formation of a durable shrunken state called a Tun. In cryptobiotic state, tardigrades can survive for many years. There are several types of cryptobiosis: **a)** anoxybiosis, a biological response triggered in deficient oxygen, **b)** cryobiosis, a biological response to freezing temperatures, **c)** osmobiosis, a reaction to excessive salinity, and the best-known state of cryptobiosis, **d)** anhydrobiosis, a response triggered by increase of solute concentration [0].In the anhydrobiotic state, the metabolic activity of tardigrades drops to a very low level[0]. This dormant state can occur at the embryonic stage as well as in adults and can be repeated multiple times. Anhydrobiosis helps tardigrade to survive in scarcity of water, but also to a number oflethal conditions such as high temperature, radiation or different kinds of chemicals, such as ethanol, hydrogen sulphide and carbon dioxide [000].Not every Tardigrade species show equal level of resistance to drying out, as there is different level of tolerance even between populations of the same species [00]. Theanhydrobiosis state is preceded by an initial phase, during which the tardigrade body undergoes a several metabolic and anatomical change that are necessary to survive the extreme conditions. The tardigrade undergoes a 'tun' state where they appear like a lifeless ball[0]. The tun form decreases the surface for evaporation and thus slows down the loss of water and the transpiration is reduced by about 50 % [0]. The tun state also prevents the damage of the internal and external organs during the drying process [0]. When all free water evaporates from the tardigrade body, it begins the process of replacing the water bound to macromolecules. The lost water is replaced with bioprotectants such as trehalose, which protects macromolecules, such as nucleic acids and proteins, from losing their proper structure[20] [10].The disaccharide trehalose plays a major role in desiccation tolerance in tardigrades [0]. Trehalose is essential for cryptobiotic organisms to survive desiccation and is thought to protect organisms by glassifying their cellular contents. Not all the cryptobiotic animal shows the trehalose secretion. However, trehalose secretion in tardigrade is still unclear [0]

This special feature of tardigrade to undergo in cryptobiosis attracting many researchers. Researchers making efforts to make this mechanism of tardigrade applicable to save humans and plants' lives. 'trehalose' is a special type of protein that has been found in cryptobiotic animals and this protein is the key of survival. It's amazing that these animals have the power to holds its life in frozen ice crystal for 30 years, previously it was thought all the tardigrades species possess trehalose but later it is proven that after being in a frozen state a few species only showed a significant amount of trehalose but few did not show the presence of trehalose at all [0,0].

During the high desiccation, TDP genes are expressed by observing this noble mechanism of trehalose

protein and TDP genes. Researchers making tardigrade economically applicable, from as a space research model to every possible medical field. Trehalose can be found in extremophiles organisms including tardigrades. The unique feature of this sugar is the ability to preserve biological molecules. There are multiple opportunities to use them for the betterment of human beings. One of the big applications of the tardigrades is "dry vaccine" implementation of these dry vaccines for covid-19, will expand its availability and will reduce dependability on cold storage, it will be increasing the shelf life of vaccines, and make every vaccine reach needy in a live condition. There are other multiple biotechnological uses of Trehalose sugar throughout the word.

This peculiar protein of tardigrade can make advancement in medical research, as tardigrades are easily available, we can make vaccine and other medicinal products or biological products at very low cost and the utmost we can save human lives using their proteins. making a vaccine or using their protein in medical fields and another ecological field at very low cost and making its access to almost anyone to every rural area and serving tardigrades to the benefits of humanity and as well as serving to all living being is our main motto.

The objectives of this review research, is to provide an overview regarding the application and economic aspects of the tardigrades and the possibilities uses of it in near future.

II. Methodology

Literature available in the public domain has been referred for the present review. Few eminent zoologists have been interviewed to achieve the objectives. We have cited the recent research paper and concluded this review paper.

III. Discussion

Tardigrade in cancer research

Fitness and adaptation were always the key for survivability. This small creature has all the ability to tolerate the lethal condition. Tardigrades can maintain their genomic integrity [0] hence their peculiar property of DNA repair and protection of DNA damage has been stealing the interest of cancer researchers as well those who are studying the effect of body ageing [0]. Transfer of DSUP gene to human embryonic kidney cell can increase the tolerance against radiation so eventually, it will be benefited for those who are undergoing radiation therapy.[0]

Dry vaccine

TDP Protein (tardigrade disordered protein) are only confined to tardigrades and it's formed by the combination of three protein viz CAHS, SAHS, and MAHS proteins [0]. TDP protein protects the cellular damage in tardigrades. Above all our motto is not to confine this protein to the books but to make this applicable in the real world, such as a dry vaccine, pharmaceutical material, or biologic material. CAHS protein creates a 3-D gel network to protect the cell we can apply this protein for the dry vaccine to get rid of the 'cold chain' method. As vaccine need a refrigerator or freezer to keep it in a continuous cold state [0] and It Create economic burden to transport or shipment the vaccine in rural areas, new developmental areas or areas which have lack of facilities, the world health organization estimates that of all the vaccine produced globally, 50% gets wasted [0] due to the cold chain method by using this protein one doesn't need to follow-up cold chain method also this method can reduce the cost of vaccines as tardigrades are easily available and this technology can eliminate the costs of freezers and refrigerators. One of the recent use of vaccines we can make covid-19 vaccines in a dry condition it's not only to get rid of the cold chain method but to increase the shelf life of vaccine and minimize the wastage Hence this can make vaccine accessible also for those who are deprived of money.

Radiation resistance

Have conflicts with radiation for ages but tardigrade has solved that too. DSUP protein protects the DNA degradation in tardigrades [0] [0] by protecting them from ionization radiation or hydroxyl peroxide treatment that generates hydroxyl radiations. DSUP protein can increase DNA damage protection by binding with human cells. Most importantly it functions normally. DSUP can increase the radiation tolerance of human cell by 40%-50% as compared to normal cells. Now out of the lab, it can implement for astronauts. As astronauts would gain less radiation in space. making the colony to the other planets is one of the issues because of their atmosphere and radiation of the sun. if human cells would act the same as the tardigrade then it would be the next step towards the future.[32]

Cryopreservation

Trehalose is one of the sugars that has been found in tardigrades [0]. Trehalose sugar can be applicable in transplantology or organ preservation [0]. This can be also significant for those women who face ovarian dysfunction due to cancer therapies. Ali eroglu successfully done cryopreservation of mouse oocytes by using trehalose sugar. He injects trehalose into the eggs of mouse and cooled them to liquid nitrogen temperature, later

exposed them to sperm. They reproduce healthy babies at a similar rate to unfrozen control [0]. The trehalose has also been successfully used to cryopreserve human pancreatic endocrine tissue. [0]

Ophthalmology

Trehalose is a disaccharide sugar. Trehalose has been found in organisms which are the survivor of extremities desiccation and anhydrobiosis including tardigrades [0]. Studies have been showing that trehalose is not effective in the minor organism but in the mammalian corneal eye [0]. This was lab experimented on murine dry eye. Mice were exposed to controlled to low humidity airflow and temperature for 21 days, this results in severe evaporative dry eye syndrome similar to the human eye. The dry eye environment results in apoptosis on the ocular surface. Further, the eye was later exposed to trehalose solution for 14 days this results in significant improvement on the ocular surface as well as a decrease in the number of apoptosis cells. This research suggests that trehalose may be worth in future research for the ocular surface in dry eye syndrome in humans [0]

Organ transplantation

Researchers has been searching for the lifesaving secrets of tardigrades. The shortage of organ donors and long transportation often required an effective preservation methodology. Similarly making available fresh organs at very short notice is still looks a difficult task.

So, the new organ preservation solution was developed at Kyoto University [0] [0]. It was made by using trehalose solution and named them ET-Koyoto and it was successfully used in lung transplantation, in fact, it showed a better result in comparison with Euro-collins which is a primary solution for lung transplantation. [0]

Pharmaceutical application

The tardigrade has a significant property of undergoing cryptobiosis. This is possible because of the trehalose sugar. Trehalose now becomes the economic interest of pharmaceutical industries [0]. Tardigrade produces trehalose sugar prior to the stages of cryptobiosis and anhydrobiosis. Trehalose protects the cellular membrane during desiccation and freezing hence this property of the sugar can be applicable in making biological products. [0] [0]

Tardigrades as bio-indicator

Environmental stress such as the level of pollution and sewage water can be detected by tardigrades. Mostly terrestrial tardigrades dwell on moss. An increase of heavy metal content in moss can reduce the number of tardigrades and specimens in moss. Tardigrades can serve as a potential bio-indicator to show the effect of air pollution on abundance of water bears [44] [0]. Also, tardigrades can be used as a bio-indicator in biological wastewater treatment. Studies show that the abundance of tardigrades was highest with the highest pollution level and a higher number of suspended solids. T. ruffoi is subjected to has a high potential to be used as a bio-indicator of nutrient load changes. [0]

Tardigrade DSUP gene enhances stress tolerance in plants

DNA damage is one of the most disgusting events in living organisms. After successfully suppressing the damage in the human cell now tardigrades are headed to save the plants. DSUP is a unique chromatin-associating protein that contributes to the organism's exceptional tolerance to harsh environmental stress [0] [0]. Justin Kirke and the team performed experimental research on tobacco plants to demonstrate the effect of the DSUP gene on plants. They expose the plant to X-ray radiation, UV-C and bleomycin, they have observed compared to the control plant, DSUP plants appeared more protected from harmful radiations. Overall studies demonstrated that the DSUP gene has expressed enhanced stress tolerance in plants. From this research, we can conclude, tardigrades can be implemented for plants in increasing tolerance to a stressful environment for future climate changes and human space settlement. [0]

Future prospective

Recently a group of scientists has identified the key protein in the tardigrades which helps them to enter in the frozen state and again to rehydrate back in life form known as" cryptobiosis". It has been proved from the experiments that a protein called "Tardigrade-specific Intrinsically Disordered Protein" (TDPs) is essential for desiccation tolerance. This protein is also called "Intrinsic disordered protein" or (IDP) [0] [0]. TDP is responsible for the formation of glass-like structures in the tardigrades. The protein helps the tardigrades in gradual drying and again to bring back their alive form by rehydrating [49]. One important advantage is that when this exact protein is separated and expressed in the human body, they still show the same properties of the protein. E.g. when the tardigrade protein genes were expressed in the human HeLa cell line, then the human DNA showed the same property of extreme tolerance against radiation. In the future there may be an experiment done by inserting the tardigrade gene into the human cell to help humans survive in extreme condition [0]. TDP gene can also help in creating a crop that can tolerate desiccation and drought. Tardigrades can be implemented for plants in increasing the tolerance to stressful environments for future climate changes and space settlement [0]. In the field of

ophthalmology trehalose may be worthy in future research for the ocular surface in dry eye syndrome in humans and successfully used in organ transplant solution at Kyoto university [0] [0] [0]

IV. Observation

A tardigrade belongs to sister group of Arthropoda they are micrometazoan and occupy diversity of niches in freshwater, marine, and terrestrial habitats [0] and Since discovery more than hundreds of research papers published on this animal. It was observed that tardigrades are among the most resilient animals due to their physiology and protein secretions. Protein secretions are very useful to human life. Now, researchers at the University of North Carolina have figured out the secret behind another of their incredible abilities-the capacity to survive for more than a decade without water [0]. It has been observed that when water bears are dehydrated, the cytoplasm in their cell turns into glass locking biological molecules in place to prevent them from becoming altered or damaged. And it all happens because of 'tardigrade-specific intrinsically disordered proteins (TDPs). Inside the cells of dehydrated tardigrades, TDP replaces the water. This forms a glass-like substance that keeps the cell structure intact. [28].

Finding life on another planet is quite attractive to astrobiologist. Scientists are not only looking for life on another planet but they are also trying to find out next home for the tardigrade.[0] [0] Considering the six parameters such as radius, density, escape velocity, revolution period, surface temperature, and surface pressure of the considered planet, a scientist has concluded that Mars could be the only planet that could be more suitable for tardigrades than other considered exoplanets. [0]

In fact, water bears could survive after humanity is long gone, researchers found. Scientists from Harvard and Oxford universities looked at the probabilities of certain astronomical events- earth-pummelling asteroids, nearby supernova blasts, and gamma-ray bursts, to name a few – over the next billions of years [0] [0] [0] [0] [0]. Then they said such catastrophic events would likely to vanish human lives,but shockingly little tardigrade has a potential to survive most of them, [0]

There is no denying that tardigrades are one of the toughest animals on our planet and are the most unique extremophiles group. Ever since it was proven that tardigrades have high resistance to the different kinds of stress factors associated with cosmic journeys, combined with their relatively complex structure and their relative ease of observation, they have become a perfect model organism for space research. This could enable researchers to determine whether taradigrade can survive and live on other planets in the solar system or on their moons. We should also continue studies on tardigrade resistance to the combined stress effect of cosmic radiation and microgravity, or low temperature and the presence of harmful chemicals. Such studies would help determine the limits of survival of the earth's multicellular organisms. This is very interesting especially in the context of searching for life on other planets and moons. [0]

Tardigrades were also sent to the moon by the Israel Bersheet's mission. The beresheet lunar lander carried thousands of books, DNA samples, and a few thousand waters bear to the moon. Surprisingly they survived in such a harsh environment by undergoing the dormant stage [0]

A new study suggests that tardigrade have a natural shield against the UV radiation. They shield itself by emitting harmless blue light but this type of resistance mechanism is not seen in most of the other species. Fluorescent in a tardigrade is linked to the radiation tolerance Researcher exposed paramacrobiotu at high level UV radiation that would have lethal to any organism for 15 minutes. Same experiment was tested on another species of tardigrade Hypsibiusexemplaris which is UV sensitive got died withing 24 hours during the experiment. tardigrade species paramacrobiotus glow brightly when exposed to UV light. Researcher also test experiment on non-florescent species i.e. H. exemplaris and C.elegance by injecting fluorescent component, surprisingly they showed partial tolerance to UV radiation. This fluorescent can help tardigrade to survive in habitats which is exposed to sun. these UV resistances can also be able aid the tardigrades in tropical region as well as the environment with high UV index. [0]

Table 1 Different types and their level of tolerance in anhydrobiosis and cryptobiosis

Different tolerance	Tolerance level
During anhydrobiosis and cryptobiosis	
Temperature tolerance	from −272.8 °C [0]to about 150 °C (up to 15 min) [0].
Liquid air Liquid nitrogen Liquid helium	Ca. -190°C [0] Ca. -253°C Ca. -272°C
Low and high atmospheric pressure tolerance	200 to 280 hPA to 7,500 MPa [0] [0]
Ionizing radiation and X-rays tolerance	Ca. 5000 GY [0] [0]
Ultra violet radiation tolerance	Between 75 and 88 KJ m2 [0]
During active state	
Temperature tolerance	38°C [0] [0]and -196°C [0]
High atmospheric pressure tolerance	Up to 100 MPa [0]
Ultra violet radiation tolerance	Between 75 and 88 KJ m2 [0] [0] [0]

Tardigrades undergoes almost dehydration by entering an arrested state known as anhydrobiosis, which allows them to tolerate ionic radiation, extreme temperature and intense pressure. Other studies demonstrated that some species inhabiting the Arctic soil can survive up to six years (74 months) at −80 °C[0] [0]. Water bears are resistant to physical stressors as well as some chemical stressors such as hydrogen sulphide, carbon dioxide, ethanol (for ca. 10 min) and 1-hexanol [0][0][0]

V. Conclusion

The tardigrade is yet not fully explored and exploited. Authors believe that tardigrade will not only be able to boost the vaccination campaigning of COVID-19 but also used as bioindicator. Many researchers also proved that protein derived from Tardigrade can help in organ donation, organ transplantation, space settlement, plant life, etc. The ability of tardigrades to survive extreme temperature opens doors for a number of advanced applications of these organisms. Tardigrades can be used as pioneer species to inhabit new ecosystems. Once tardigrades inhabit these ecosystems, other invertebrates might also move towards them along with the predators of tardigrades. The proteins of tardigrades that protect them against such conditions can be used in crops and vegetation so that they can survive severe, long-lasting droughts. Similarly, the use of these proteins in medication might allow the medicines to be store at room temperature instead of having to be chilled. This process will enable the transport of various medications and vaccines to remote areas that might not have the facility of freezers. Because they can withstand the conditions of outer space, tardigrades are considered as the model organisms for space research. Tardigrades can be used in studies related to extreme environments and extreme physical conditions. Their cells contain special proteins that protect DNA from breaking down under stress, such as extreme heat, cold or radiation. In 2007, tardigrades were exposed to the radiation of space for 10 days and survived. Researchers sought to impart their survivability in humans by binding tardigrade proteins to human cells. The produced cells showed 40-50% reduced x-ray damage

compared to normal human cells. If these results could be replicated outside the lab, astronauts would gain improved protection against the radiation of space.

DARPA created the Biostasis program to support research in slowing the body's metabolic processes following traumatic injury. Based on the tardigrade's ability to dehydrate and suspend their bodily processes for decades, applying this state to a human being could save them from bleeding out or stop the progression of sepsis or damage from a stroke or heart attack.

Tardigrades can be applicable in every possible field. We need to more understand the tardigrades from the biological aspects to implement them from 'Survivor' to 'savior'. Trehalose protein can be used in numerous ways from an economical point of view. Tardigrades protein stealing the interest of pharmaceuticals industries because they contain high potential in making biological products and their availability in every corner of the earth can make the products low cost. We can preserve the vaccines or we can use them as dry vaccines for the most recent crises "covid" as well as organ for transplantation. We can serve tardigrades for the women who are suffering from ovarian dysfunction due to radiation therapy. Tardigrades can not only use in the medical and pharmaceuticals field but as well we can increase the stress tolerance in plants and humans against the radiation for the future prospects of space settlement

Though there are studies emphasizing their identification and importance, there is a need for more research to understand their relevance in different fields of science. Above all this application, using a tiny creature protein to such a big complex human body is still a big task to the researchers.

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