

I can Eat Uranium-My Name is Deinococcusradiodurans

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I was discovered way back in 1956 in a can of meat product, which was treated with large dose of radiation to remove hazardous bacteria. But I could sustain this treatment. That was when the world came to know about a bacterial species *Deinococcus* sp. Myself *Deinococcusradiodurans* (DR1), a pink colour, aerobic, tetrad-shaped bacteria. Since the discovery, I have fascinated the scientific world with peculiar properties and applications. The more you explore, the more I excite you. We are the only group of bacteria that holds a place in *Guinness Book of World Records*, as “the world’s toughest organism”. Are you excited to know what is so special about us? Let me explain, I can tolerate up to 5000 Gy (Gray denoted as Gy is a derived unit of ionizing radiation dose in the International System of Units (SI)) of ionizing radiation and withstand nutrient starvation. We can live in vacuum for up to six weeks, stay dehydrated and live through fluctuations in pH. If you are wondering what is so special about this, for your information, 100 Gy of ionizing radiation can kill a human. These unique features make me stand out from other bacterial species. The world calls us the tough bacteria, indeed a very apt name! To add, we are not hazardous and do not cause any disease.

Although I differ from other bacteria in many ways, we lack the ability to form a biofilm. Wait, but what is a biofilm? Let us understand the term with a story, once there was an old man, who wanted to teach his three selfish sons a lesson. He got a bundle of sticks and asked his sons one by one to break the bundle. No one was able to break the bundle. But when he untied the bundle

* Ms. Manobala T, Ph.D. Scholar from Department of Applied Science & Technology, Anna University, Chennai, is pursuing her research on “Studies on *Deinococcus Radiodurans* R1 Biofilm formation and its Implications in Bioremediation.” Her popular science story entitled “I can Eat Uranium-My Name is *Deinococcus Radiodurans*” has been selected for AWSAR Award.

and asked them to break the sticks, they were able to do it easily. The moral of the story is united we stand, divided we fall. The old man thought a big lesson to his sons about the art of living in unity. Probably, humans may fail to know the importance of being in unison, but bacteria do not let it happen. Scientifically, biofilm can be defined as any group of microorganisms in which bacterial cells stick (aggregate) to each other and often to a surface. These adherent cells become embedded within a slimy matrix that is composed of extracellular polymeric substances (EPS). This polymeric extracellular matrix constitutes polysaccharides, proteins, lipids and DNA. These components help bacteria to adhere to a surface, sustain its life and replicate. In natural environment, almost 99 per cent of all microorganisms live as surface attached communities known as biofilms.

Do you know that the scalp that a dentist removes while scaling is a biofilm? But these biofilms are not required for any particular purpose (they are the ones that cause bad breath!) and they sometimes become harmful to us. The slime layer that forms in a water pipeline is another example. Biofilm formation is not restricted to a single species, it may sometimes comprise multiple bacterial species happily living together. Also fungi form biofilm. When the bacteria exist in the form of biofilm, they possess a lot of advantage. They become more robust and tolerate different environmental conditions. Do these properties have any relevance for the human use? Could this biofilm be used for environmental remediation applications? The effective usage of biofilm for bioremediation (bioremediation is defined as usage of microorganisms to break down pollutants) purpose has potential environmental clean-up applications. Use of biofilms for environmental remediation purpose has potential advantages than their planktonic counterparts.

As I have mentioned, I do, not have the innate ability to form a biofilm. A joint venture project by Anna University, Chennai and Bhabha Atomic Research Centre facilities, Kalpakkam has been successfully executed by a doctoral student Manobala. A genetically modified *Deinococcus radiodurans* R1 (denoted as DR1_bf+) has shown a biofilm forming ability. Hooray!! This is the first time that I have been able to form a biofilm, Yes, it is the first report. Thanks to this new study. It has been a completely different experience for me. I was able to adhere to the surface and form biofilm in the presence of calcium ions and an antibiotic kanamycin. Initial studies with this biofilm have confirmed the role of outer cell wall proteins for the initial adhesion. A surface layer protein (*slp*) expression is comparatively increased in genetically modified DR1_bf+. A comparative protein analysis of DR1 and DR1_bf+ has confirmed the involvement of outer layer proteins in biofilm formation behaviour. And now I am in a great zeal that I am able to form biofilm, which we could not do earlier. The genetic modification by addition of *gfp* gene and kanamycin resistance marker has serendipitously given me this capability.

Major outcomes

I was wondering, how would this biofilm forming ability affect me. I am happy that there was no alteration in my metabolic abilities and characteristics. To my surprise, in the biofilm mode I was able to tolerate up to 1000 mg/L of uranium, which I could not do when I was alone. Because of high radiation resistance tolerance and also biofilm forming ability, I have potential application in

bioremediation. Initial studies were done using uranium. Our bioremediation results showed that DR1-bf+ biofilm had significant capability to remove uranium that too at very high rate which was ~75 per cent removal within 30 minutes post treatment. This observation implicates the potential of DR1_bf+ for the development of biofilm-based bioremediation process for uranium removal from radioactive aqueous waste solutions. The biofilm mode of DR1_bf+ has shown tolerance to high concentrations of uranium, up to 1000 mg/L. Biofilm could withstand very high concentrations of uranium solution, i.e. 1000 mg/L not only due to the fact that DR1 has significant heavy metal tolerance but also of the fact that biofilm mode of life reduces the toxicity to the microbes as compared to that of planktonic single cells. This DR1-bf+ biofilm-mediated uranium removal method showed significant higher efficiency in terms of both performance as well as time (75 per cent uranium removal within 30 min post treatment) as compared to its planktonic counter parts

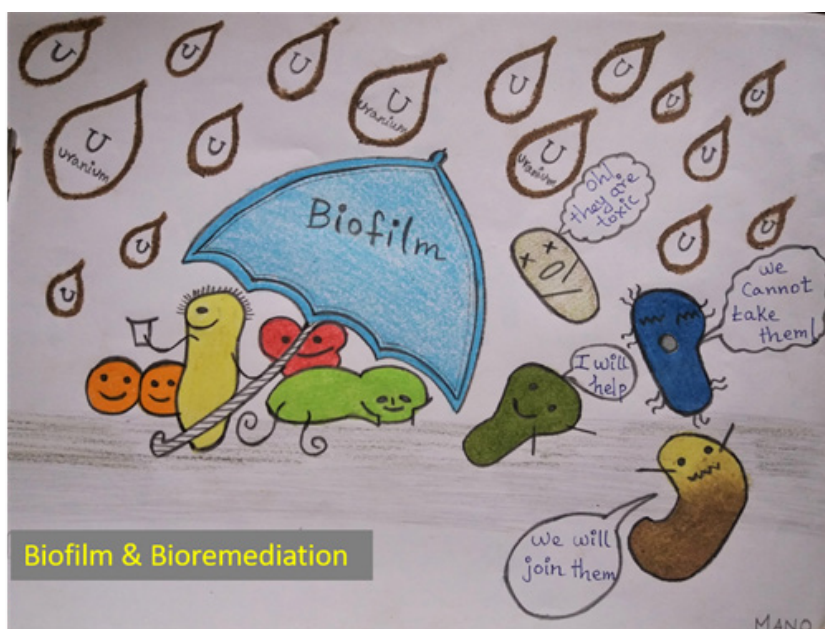


Figure 1: Cartoon representation of a biofilm and bioremediation process, where the biofilm is treated with uranium

as well as DR wild-type strain. We do not require special conditions such as maintaining anaerobic condition in this removal process and tolerance to high concentrations of uranium makes DR1_bf+ biofilm-mediated uranium removal process highly promising.

Uranium is precipitated in the form of yellow visible precipitates (figure 2). The X-ray Photoelectron Spectroscopy (XPS) studies have shown the precipitates in the form of mineral crystals. Adsorption on the surface of the bacterial biomass is the initial step followed by the precipitation of uranium.

Acid phosphatase (Acid phosphatase is an enzyme that hydrolyses organic phosphates in acidic pH) activity is also observed in my biofilm, which aids in the bioremediation of uranium.

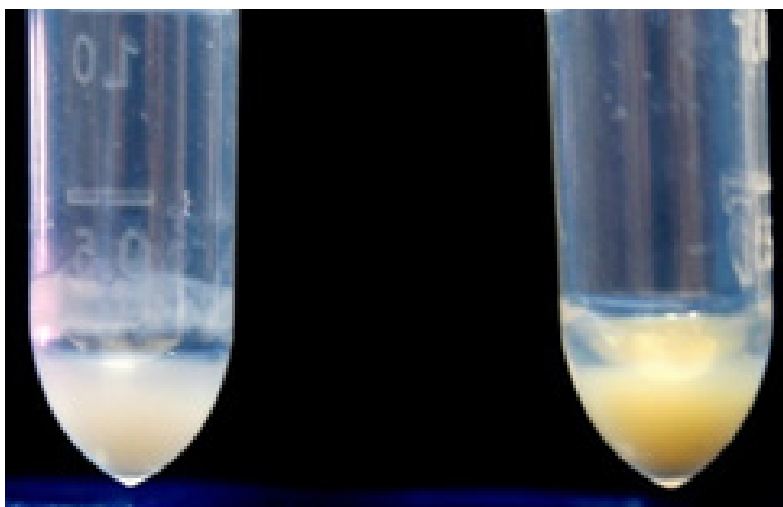


Figure 2: Scraped biofilm (a); Visible yellow precipitate observed after treating biofilm with uranium (b)

For continuous uranium removal studies, scrapped biofilm in the form of columns has been devised and has shown potential field-level applications.

Conclusion

Since DR1 wild-type bacterium does not form biofilm, a genetically modified strain of DR1-bf+ with biofilm forming capacity was tested for its potential to remove uranium from aqueous solution. It was also found that if DR1-bf+ grown in the presence of calcium, EPS production was

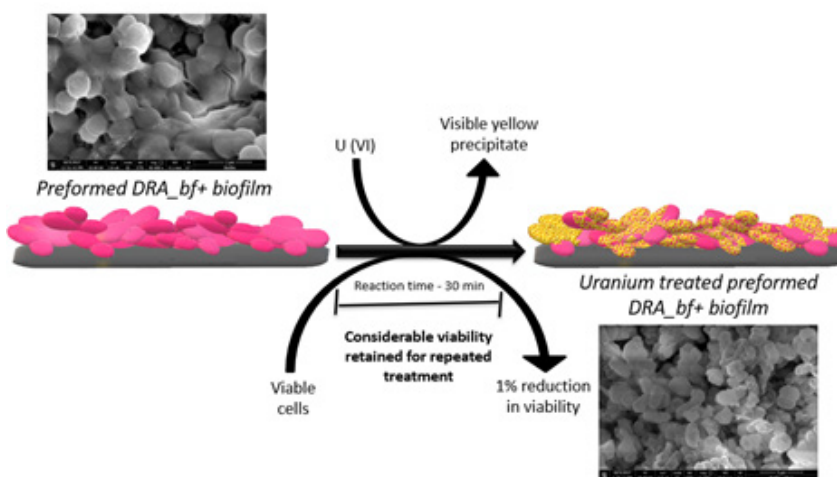


Figure 3: Pictorial representation of complete bioremediation process

significantly enhanced and it acquired extraordinary potential of uranium precipitation as compared to its planktonic counterpart. Without much change in the biofilm biomass, a proportionate age dependent increase in bioremediation process has proved the involvement of biofilm matrix whose composition changes over time. This observation implicates the potential of DR1_bf+ biofilm-based bioremediation process for uranium precipitation.

The biofilm forming capability has improved our bioremediation capability and I am sure this will help in the environmental clean-up. Hope, I will be able to help our environment get rid of the harmful contaminants.