

BIOLOGICAL DECONTAMINATION OF RADIONUCLIDES IN INDUSTRIAL AND ENVIRONMENTAL WATER USING A NEW HIGHLY RADIORESISTANT MICROALGA

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ABSTRACT

Nuclear energy technologies generate radioactive and chemically toxic compounds including mainly carbon-14 and radioactive metals. Treatment processes rely on physico-chemical methods, which, though efficient and robust, do not completely remove carbon-14 and are not adapted to large environmental volumes. Biological remediation technologies may constitute an interesting alternative to reduce radioactive releases in aqueous effluents or to clean-up accidentally contaminated water. We recently isolated a new microalga from a nuclear facility, *Coccomyxa actinobiotis*, which withstands huge ionizing radiation doses, up to 20 kGy. This microalga very efficiently and rapidly decontaminates most of the metallic radionuclides contained in nuclear effluents and 85% of carbon-14 in a few hours. The process feasibility was demonstrated at real-scale for *in situ* decontamination and pilote-scale for remote water treatment.

Keywords: biotechnology, microalga, nuclear industry, environmental water, radionuclide decontamination

1 INTRODUCTION

Nuclear energy technologies generate radioactive and chemically toxic compounds including tritium, carbon-14 and radioactive metals. They require processes for radionuclide decontamination inside the facilities themselves and of the liquid and gaseous effluents released into the environment. Treatment processes frequently used to remove radionuclides from liquid streams are based on physico-chemical methods including evaporation, solid/liquid separation by filtration, centrifugation or decantation possibly associated to a chemical precipitation/flocculation process, reverse osmosis,

ultrafiltration, sorption, and ion exchange [1,2]. These methods are efficient and robust; however, they do not completely remove some radionuclides such as carbon-14 and they would not be adapted to the treatment of large environmental volumes. Alternative technologies are thus needed to reduce radioactive releases in aqueous effluents or to clean-up accidentally contaminated water.

Biological remediation technologies, based on living organisms or extracts from living organisms, are already used in many industrial applications and often offer efficiency, cost and environmental impact benefits against conventional technologies [3]. They may constitute an interesting alternative to clean-up radioactive streams as well. However, very few bio-based technologies have been proposed in the nuclear field. For maximal decontamination performance, such technologies would require organisms that simultaneously accumulate radionuclides while sustaining their chemical and radiological toxicity. The most radiation-resistant organisms described so far are prokaryotes, including the bacterium *Deinococcus radiodurans*. But *Deinococcus radiodurans* needs genetic engineering to acquire toxic-metal resistance and remediation capabilities [4].

We recently isolated a new green microalga from a nuclear facility which combines both properties. We characterized its radioresistance and its radionuclide accumulation properties and compared, on the basis of remediation experiments performed at lab-scale and at real-scale, its decontamination efficiency and the ultimate waste volume generated to that of the usual physico-chemical treatment. This alga is an excellent candidate for new biotechnology methods of remediation of contaminated industrial or environmental water.

2 RESULTS AND DISCUSSION

2.1. Resistance to ionizing radiation

This organism, *Coccomyxa actinabiotis*, is a unicellular green microalga of ellipsoidal shape measuring about 3.8 μm x 6.8 μm (Figure 1). It contains a big parietal chloroplast with starch granules. Cells in division can be observed in Figure 1.

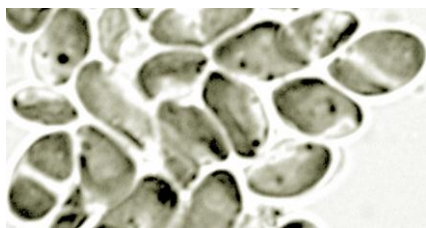


Figure 1: *Coccomyxa actinabiotis* cells.

This eukaryote was isolated from a storage pool for spent nuclear fuel elements, where it is continuously submitted to ionizing radiation. It withstands huge ionizing radiation doses, up to 20,000 Gy, when subjected to an intense γ-irradiation flux of 4,000 Gy/h [5]. The lethal dose to half the population (LD₅₀) is 10,000 Gy [6], which is comparable to that of the bacterium *Deinococcus radiodurans* and is outstanding for a eukaryote. The lethal dose corresponding to 50 % mortality after γ-irradiation for other reference species or radioresistant species is indicated in Table 1. For microalgae, this value is between 30 and 1,200 Gy [8].

Organism	Resistance to irradiation
Human	10 Gy
<i>E. coli</i> [7]	~ 200 Gy
Microalgae [8]	30-1200 Gy
<i>Chroococcidiopsis</i> [9]	2.5 kGy
<i>Pyrococcus abyssi</i>	3 kGy
<i>Coccomyxa actinabiotis</i> [6]	10 kGy
<i>Deinococcus radiodurans</i> [7]	6-10 kGy

Table 1: Comparison of resistance to ionizing radiation of various organisms (LD₅₀, for Human, LD₁₀₀).

Such high doses hardly affected the cellular metabolism of *Coccomyxa actinabiotis*. The growth after irradiation at doses of up to 6,000 Gy was not significantly different from that of the control. Nine days after irradiation, the metabolite content of the microalgae did not significantly differ compared to the non-irradiated control, whatever the irradiation dose, suggesting the complete recovery of cellular functioning [5].

2.2. Accumulation of radionuclides

Furthermore, this microalga very efficiently and rapidly accumulates most of the metallic radionuclides contained in nuclear effluents including ²³⁸U, ¹³⁷Cs, ^{110m}Ag, ⁶⁰Co, ⁵⁴Mn and ⁶⁵Zn with decontamination rates above 90% [10]. It also fixes, via metabolically active processes, mineral and organic species of ¹⁴C which is, together with tritium, the main radionuclide present in effluents issuing from nuclear facilities. More than 85% of ¹⁴CO₂ can be decontaminated in a few hours.

One of the most employed and efficient radionuclide decontamination method for water at nuclear facilities is ion-exchange on resins. Figures 2 and 3 show the decontamination efficiency of microalgae compared to that of the resins in suspension in the same nuclear effluent. When using 160 mg microalgae fresh weight *i.e.* 16 mg dry weight/100 ml nuclear effluent, the decontamination efficiency was similar to that obtained with 80 mg resin/100 ml effluent for the γ-emitter radionuclides ^{110m}Ag, ⁵⁸Co, ⁶⁰Co, and ⁵⁴Mn after a 1 h contact time (Figure 2). ⁵¹Cr decontamination by microalgae was lower. But ⁶⁵Zn decontamination by microalgae was twice higher, probably because zinc is a physiological metal incorporated by the cells via active mechanisms.

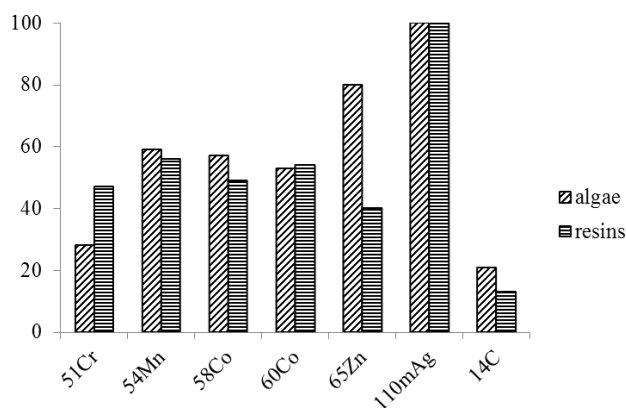


Figure 2: Comparison of decontamination efficiency of microalgae to that of ion-exchange resins after a 1 h contact time.

After a 24 h contact time, the decontamination efficiency using algae increased, though less or identical to that obtained using resins, except for ¹⁴C (Figure 3).

Decontamination of ¹⁴C is usually not fully achieved using ion-exchange resins and reached 27% in this experiment. Using microalgae, and in the light, ¹⁴C was efficiently decontaminated owing to its incorporation inside the microalgal cells through metabolically mediated processes, particularly through photosynthesis. In that kind of application, this microalga efficiently supplements the physico-chemical decontamination methods classically employed.

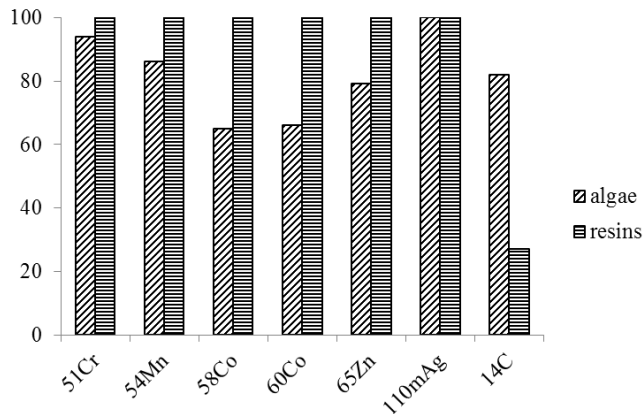


Figure 3: Comparison of decontamination efficiency of microalgae to that of ion-exchange resins after a 1 h contact time.

The microalgae volume could be reduced by 90% by drying. Once dried in the light, the algae retained the totality of the γ -emitters and 97% of the ^{14}C fixed during the decontamination step.

2.3 Real-scale decontamination test

We tested radionuclide bio-decontamination at real-scale in a 360-m³ storage pool of radioactive components by using *Coccomyxa actinabiotis* in suspension in water and comparing its decontamination efficiency to that of the conventional method, namely ion-exchange resins (Figure 4).

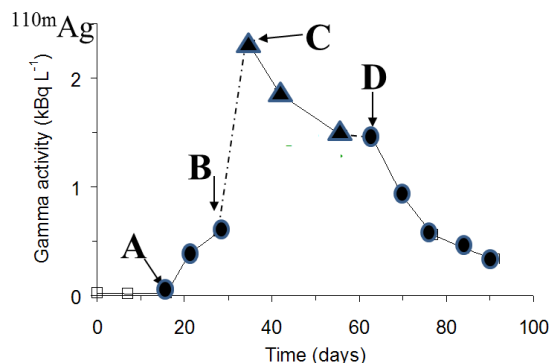


Figure 4: Decontamination of a storage pool of nuclear components using microalgae (triangles) and ion-exchange resins (circles).

Resin-based purification, used from time A to B, was stopped at time B and replaced by algae-based decontamination from time C, and for 21 days. A decrease in the main radionuclide concentration, $^{110\text{m}}\text{Ag}$, was simultaneously observed, originating from its uptake by

suspended algae, which were then collected onto filters. During that period, the algae removed 740 MBq of γ -emitters including 310 MBq $^{110\text{m}}\text{Ag}$, 270 MBq ^{51}Cr , 90 MBq ^{124}Sb , and 30 MBq ^{60}Co from the pool. The mean activity collected by *C. actinabiotis* was 20 MBq/g dry weight. About 40 g dry weight of microalgae was used in that experiment. From time D, the resins were restarted.

3 CONCLUSIONS

In summary, we have discovered a unique microalga with an exceptional radioresistance, comparable to that of the most radioresistant bacteria. This photosynthetic microorganism can survive with only water, light, CO₂, and a few dissolved minerals. It can thrive in a radioactive environment and is also capable of capturing and concentrating, rapidly and efficiently, radionuclides in nuclear facility water. The feasibility of the bio-purification of radionuclides at real-scale was demonstrated. New means of remediation are now conceivable using this micro-alga. They may be applied in nuclear facilities where they would complement conventional methods and reduce the volume of radioactive waste, in the final cleaning of water to reduce radioactive emissions into the environment, or in decontamination of accidentally polluted water.

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