

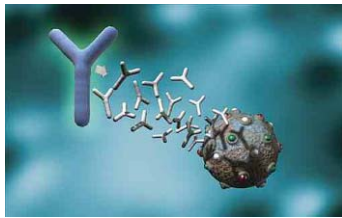
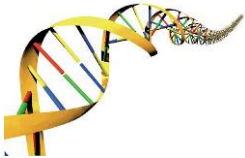
# Stakes and Limits of Bioremediation

## Bioremediation

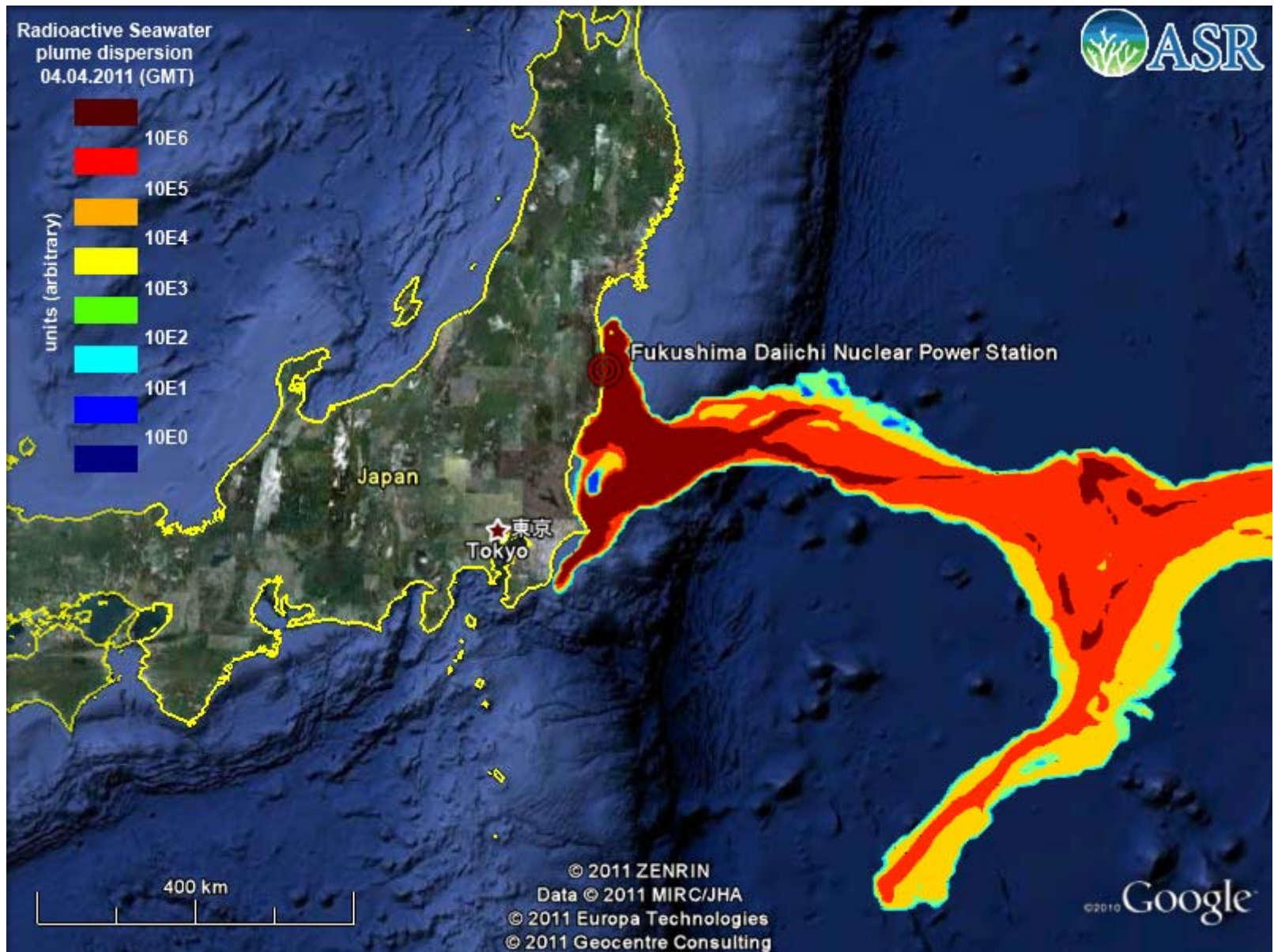
The branch of biotechnology that uses biological processes to overcome environmental problems

The use of biological agents, such as bacteria, fungi, or green plants, to remove or neutralize contaminants, as in polluted soil or water.

The use of green plants to decontaminate polluted soil or water is called **phytoremediation**.

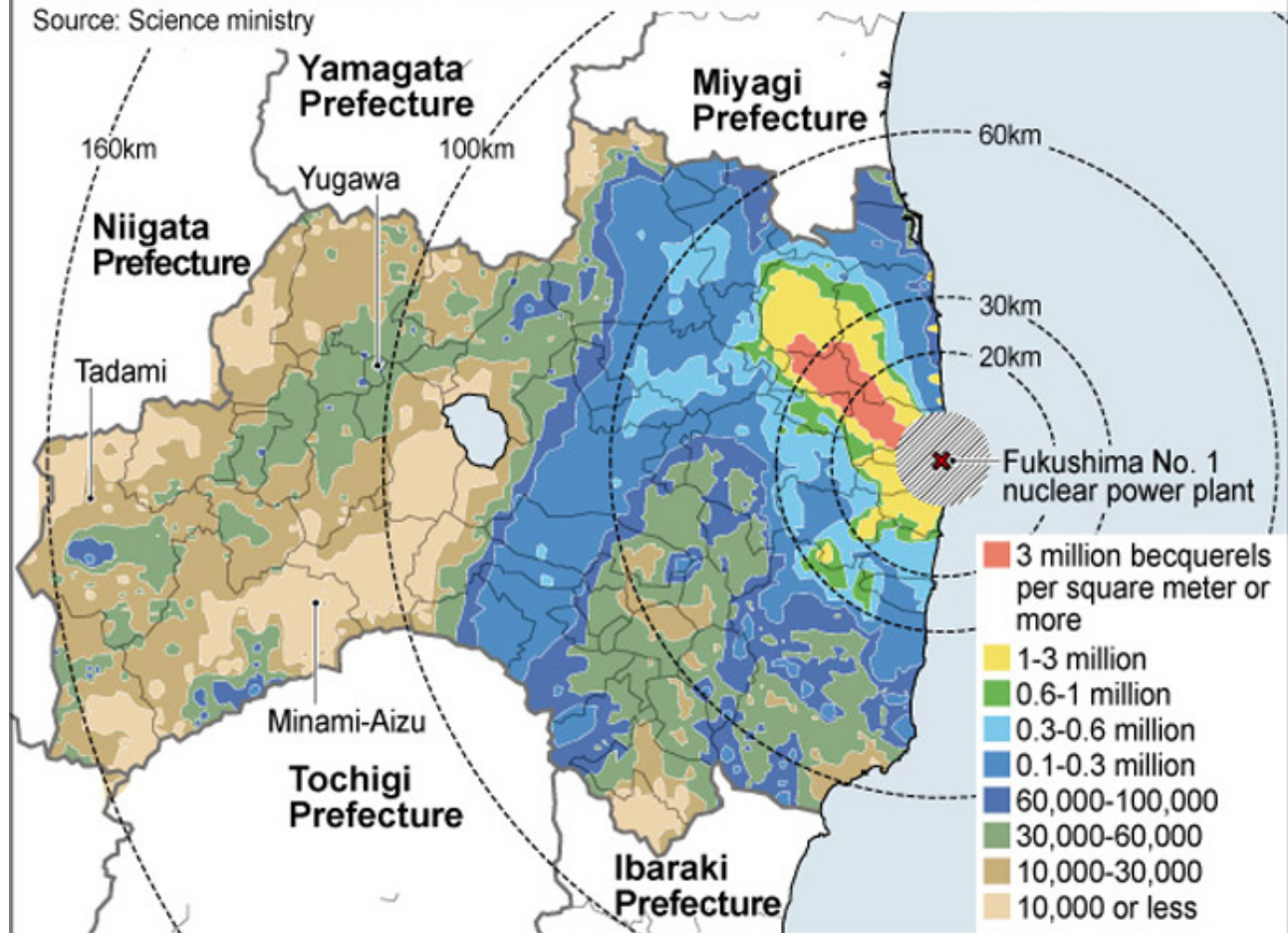


## Bioremediation and the cleanup of contaminated environments



### Cesium-134 and cesium-137 accumulations in Fukushima Prefecture

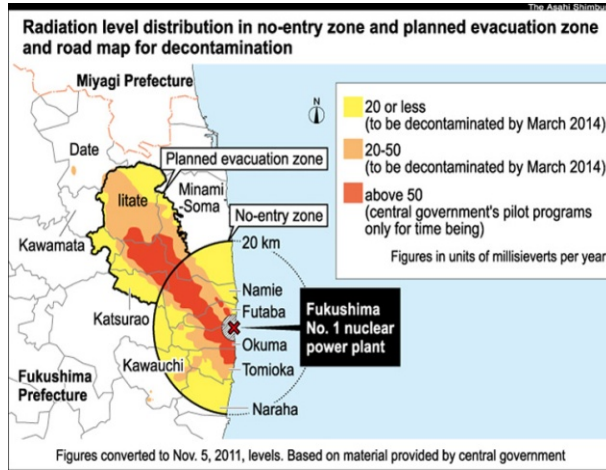
Source: Science ministry



Seuil de 5 mSv/an  
1800 km<sup>2</sup>

Seuil de 1 mSv/an  
13000 km<sup>2</sup>

## Post-Fukushima context



### First option:

Use of physico-chemical approaches for decontamination  
Removal of a soil layer (5-10 cm), deep plowing, adding K

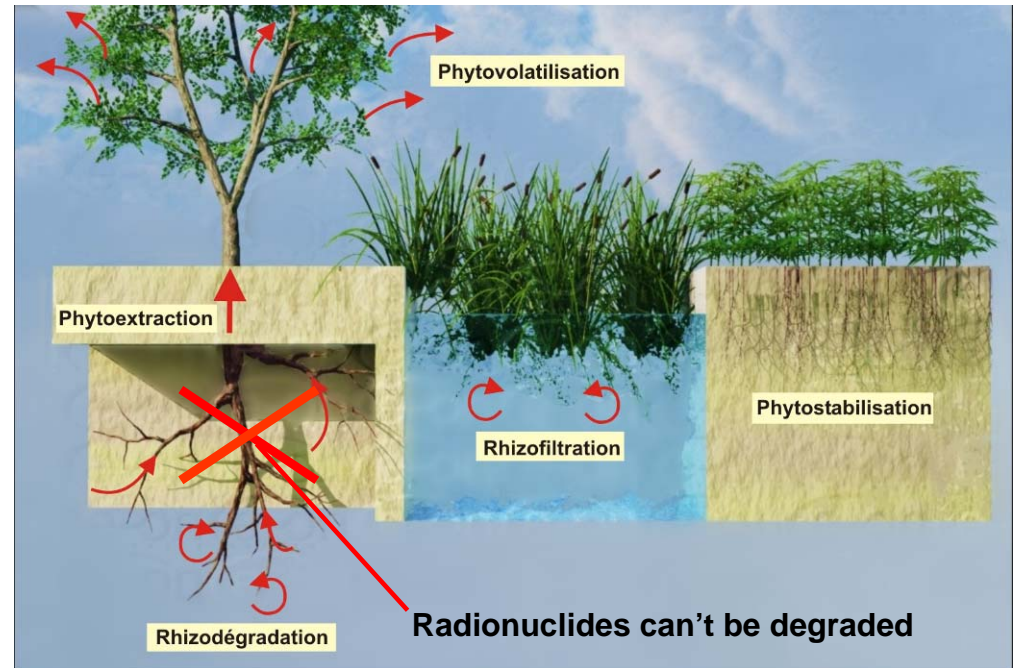
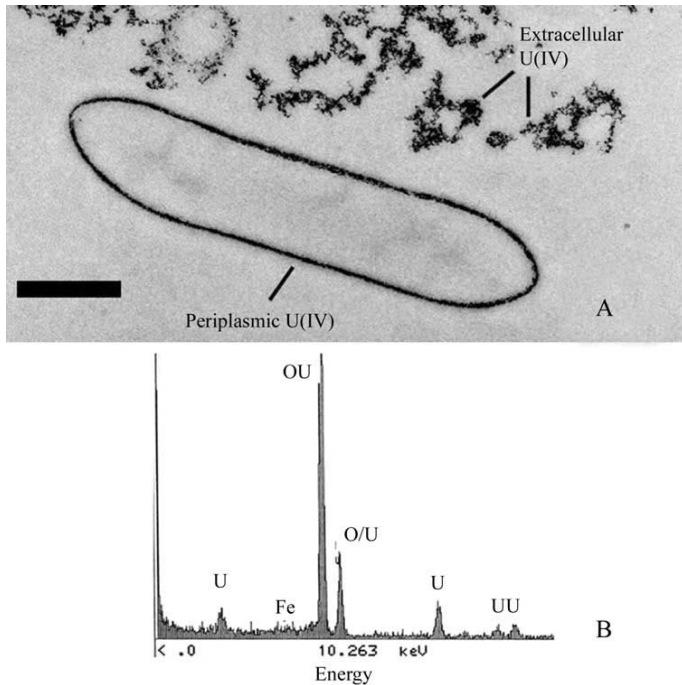
Cleanup < 6 mSv/year:  $1.8 \cdot 10^3 \text{ km}^2 \times 5 \text{ cm soil}$   
→  $10^8 \text{ m}^3$  of  $^{137/134}\text{Cs}$  contaminated waste!!

No industrial means to stock  
and treat such an amount  
of contaminated material



# Is bioremediation of radionuclides an option ?

There is no alternative for large scale remediation



Bioremediation can change their bioavailability

Phytoremediation can stabilize and extract the contamination

# Major interest of bioremediation

- Works *in situ* with a minor impact on the environment compared to physical or chemical techniques; preservation of the landscape and of soil fertility for the future

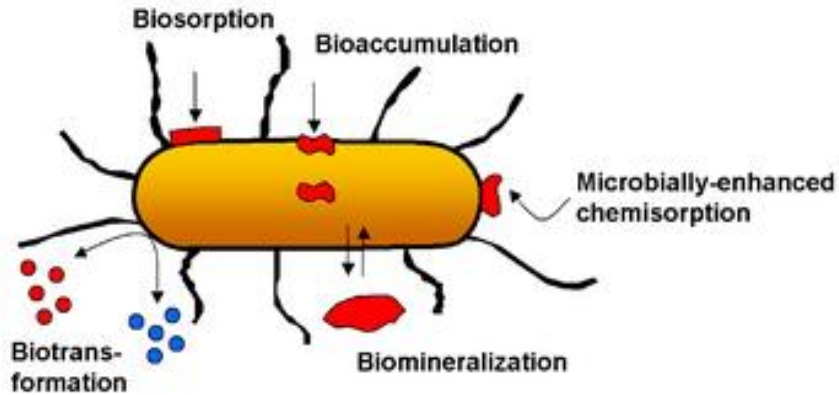


- Can remediate large areas with moderate to low contamination and decreases erosion by wind and water
- Cost is moderate, generally 10 to 100 fold lower than other techniques and extended in time
- Well accepted by the population as a “green technology”

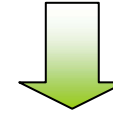


Recycling of biomass through the production of energy is economically interesting and creates an activity in areas where agronomy is impaired

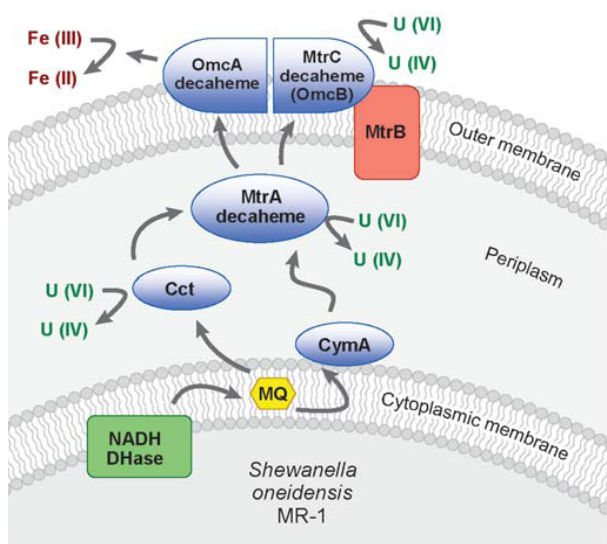
# Bioremediation using bacteria and electron transfers



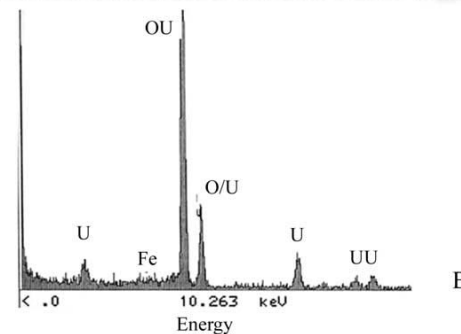
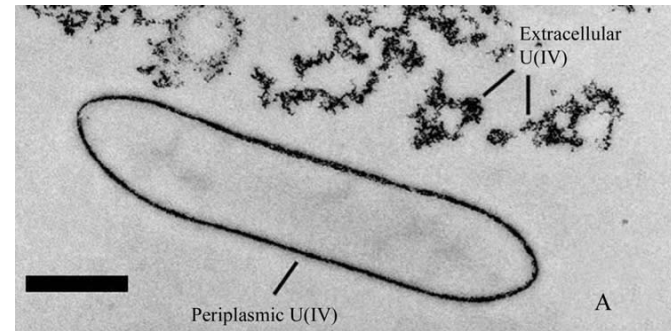
U(VI) soluble in water and toxic



U(IV) precipitation, non bioavailable



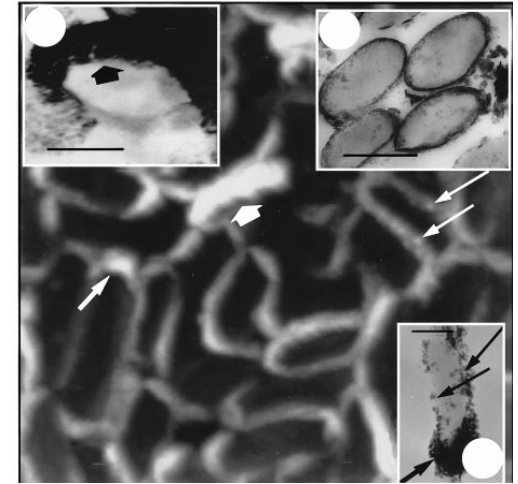
- Geobacter metallireducens*
- Shewanella oneidensis*
- Desulfotomaculum reducens*
- Thermoterrabacterium ferrireducens*



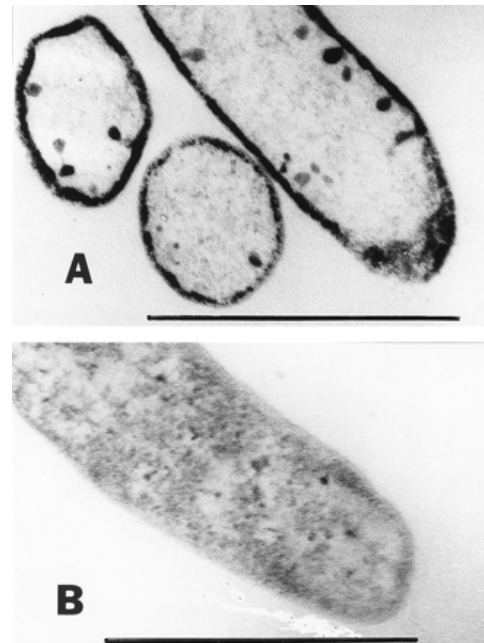
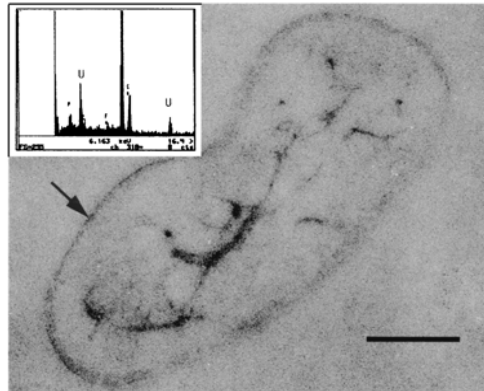
# Bioprecipitation of U, Tc, Pd, Pu... using bacteria

U bioprecipitation due to a phosphatase activity leading to the formation of  $UO_2PO_4$  complexes

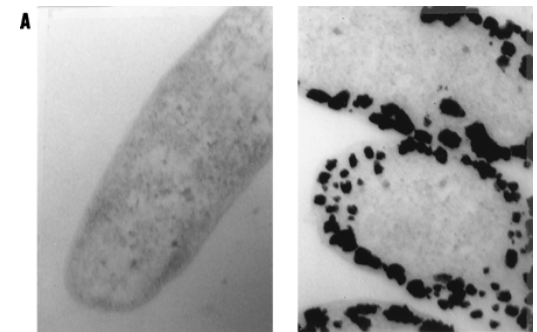
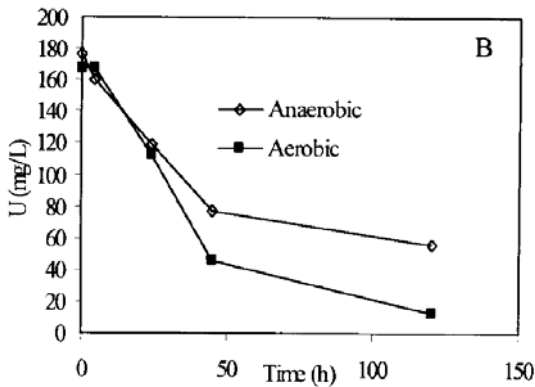
*Pseudomonas* CRB5 : granules of U in the cytoplasm associated with polyphosphates



Tc Reduction and accumulation at the cell wall of *Shewanella*

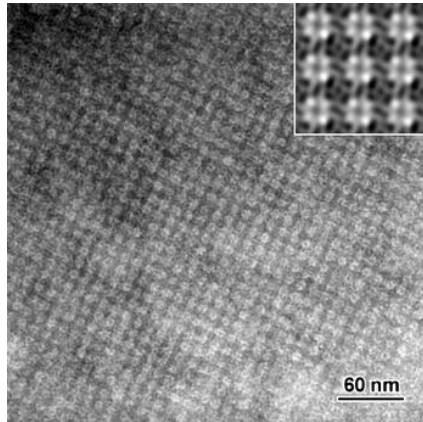


Pd accumulation in sulfate-reducing bacteria





# Bioprecipitation of U, Tc, Pu... using bacteria



Metal binding by bacteria from uranium mining waste piles and its technological applications

K. Pollman et al. (2006) *Biotechnology Advances* 24, 58– 68

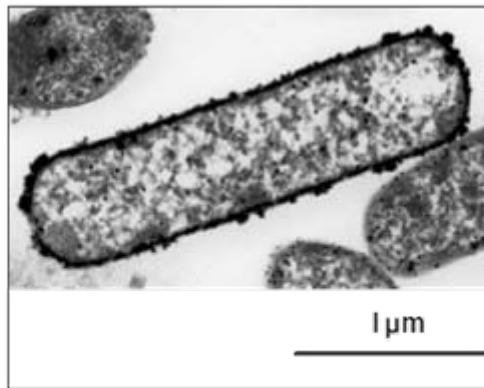


Fig. 4. TEM micrograph of Pd nanoclusters deposited on the cell surface of *Bacillus sphaericus* JG-A12.

*Bacillus sphaericus*

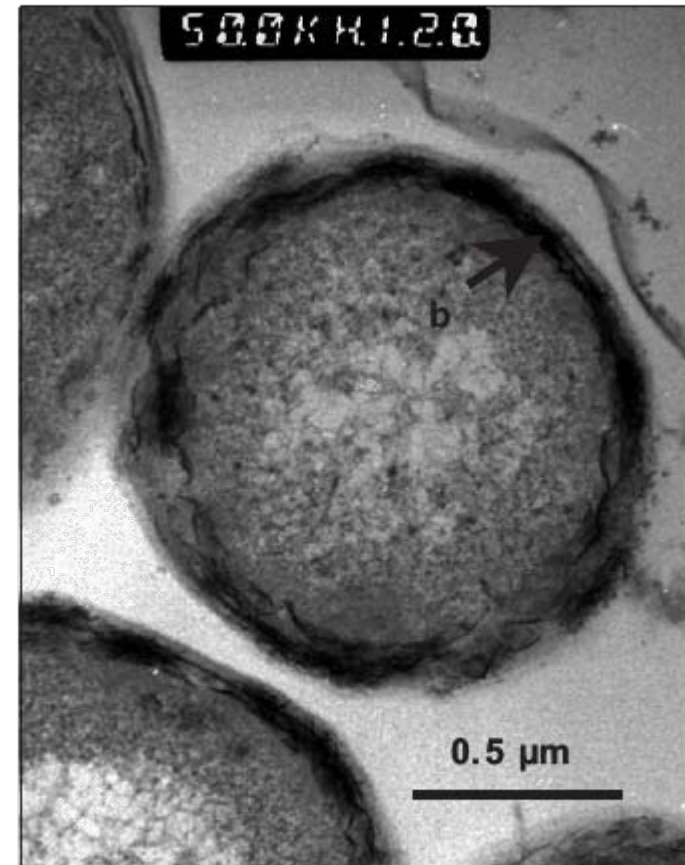


Fig. 2. TEM micrograph of uranium accumulated on the cell surface of *Bacillus sphaericus* JG-A12.

## Bacterias can be integrated in various polymers and included in a filtration chain



Composite ceramic tanks for thermophile bacteria 300 m<sup>3</sup>  
Perring, South Africa gold extraction from (arseno)pyrite

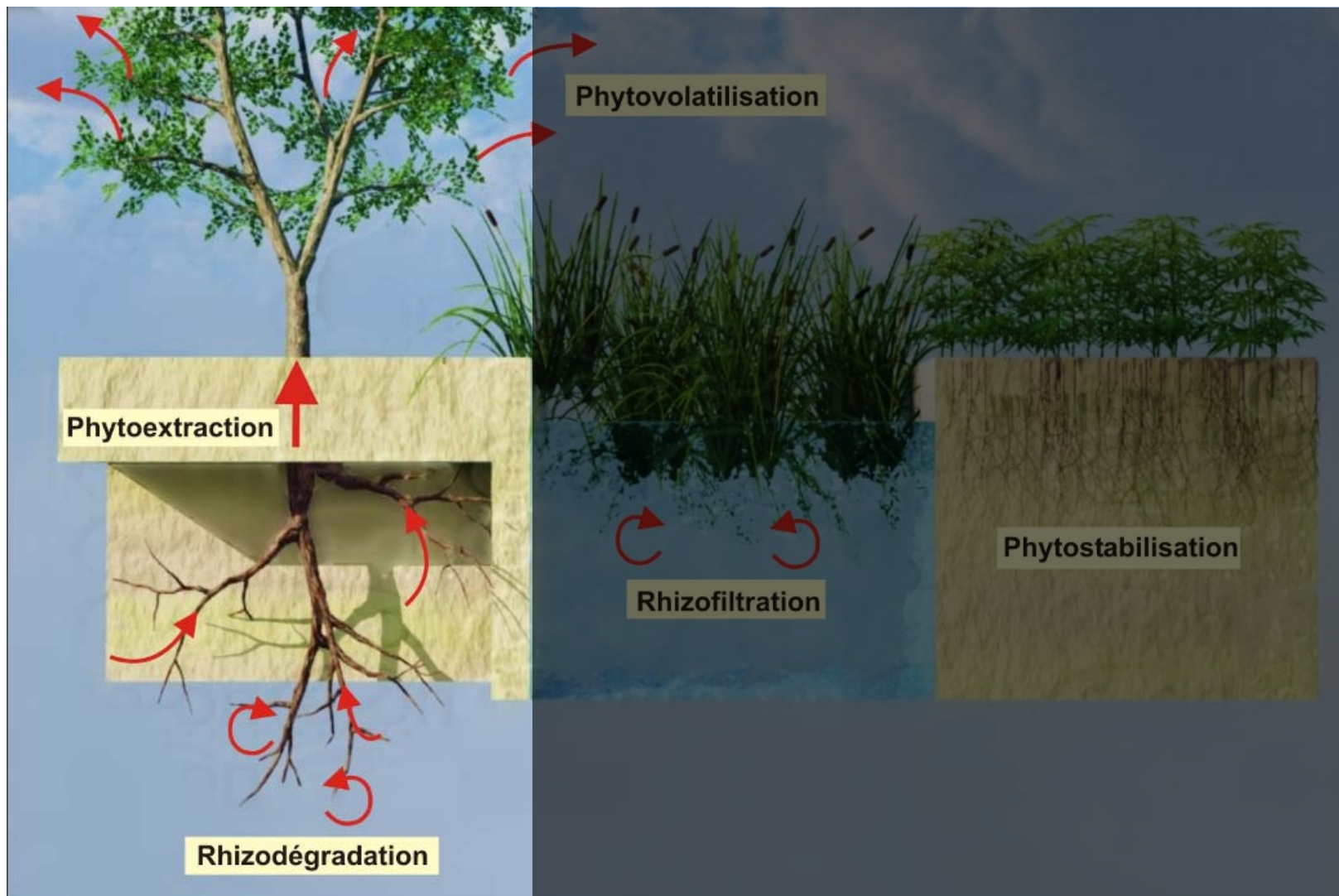
## Bacterias can also be used directly on the field for bioleaching or biostabilization

Bioleaching of U from minerals containing 0.05% à 0.15%  
 $U_3O_8$

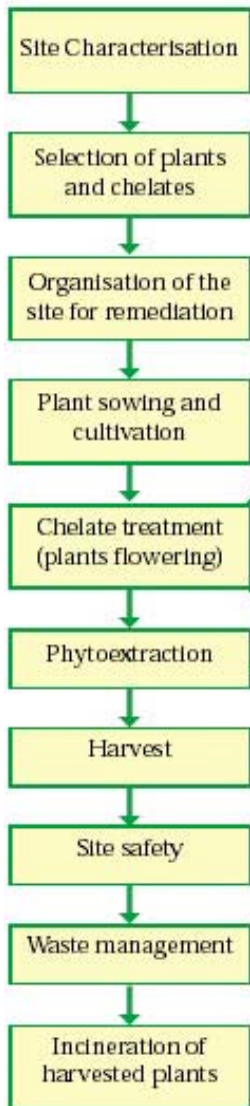


# Phytoextraction of radionuclides

$$\text{Ext} = \text{Biomass} \times \text{TC}$$

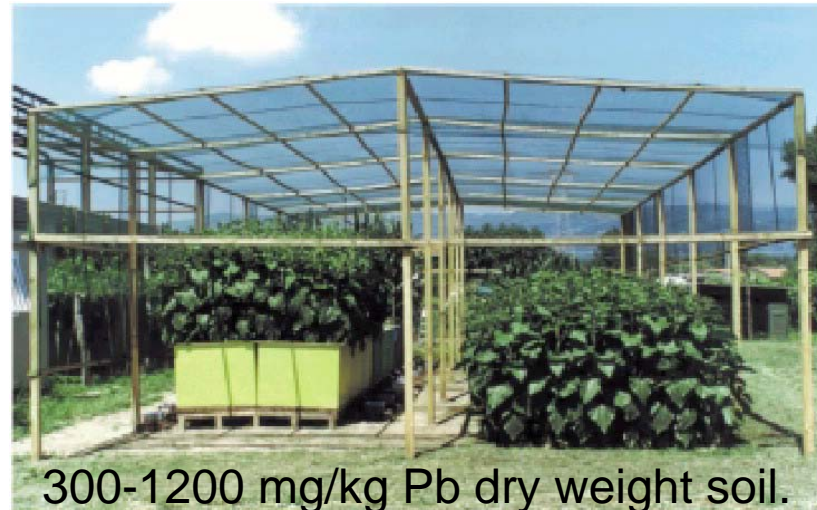


# PhyLeS Project



**Chelate treatment**

- K<sub>2</sub>EDTA: 1 and 5 mmol/kg dry weight soil
- HEDTA: 5 mmol/kg dry weight soil
- CONTROL: without chelates



	Phytoextraction coefficient (PhC)
+ K <sub>2</sub> EDTA, <i>B. juncea</i> cv. 426308	2.7
+ HEDTA, <i>B. juncea</i> cv. Chao Chow	1.78
+ K <sub>2</sub> EDTA, <i>B. juncea</i> cv. Chao Chow	0.82
+ K <sub>2</sub> EDTA, sunflower	0.34
+ HEDTA, sunflower	0.34
Control, <i>B. juncea</i> cv. 426308	0.18
Control, <i>B. juncea</i> cv. Chao Chow	0.06
Control, sunflower	0.03

↑ x90 ↓

Using *Brassica juncea* cv. 426308 and 5 mmol HEDTA/ kg d.w. soil, the estimated number of years needed to phytoremediate a soil comparable to that of our pilot system is  $\cong 20$

# Bioavailability Is essential

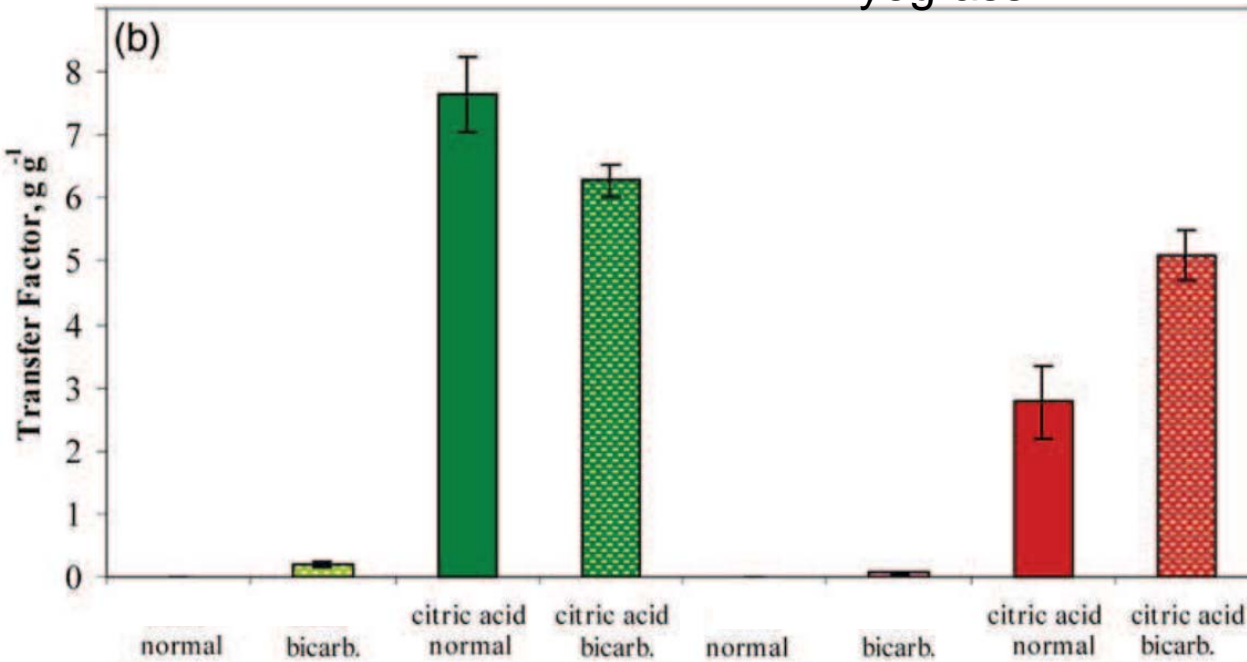
## Phytoextraction for clean-up of low-level uranium contaminated soil evaluated

H. Vandenhove\*, M. Van Hees

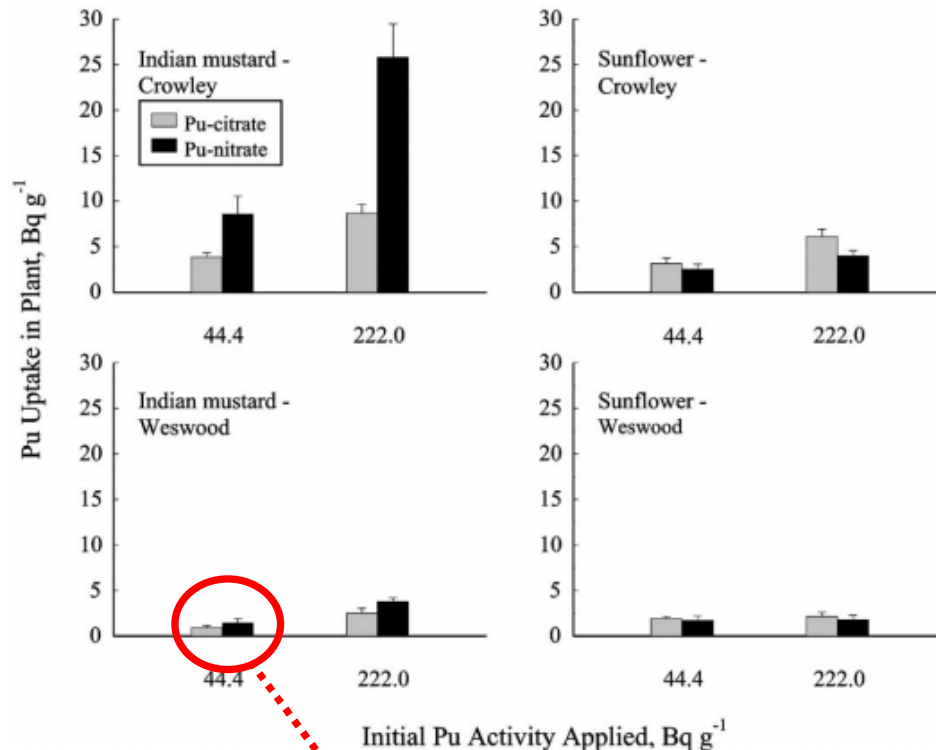
*Belgian Nuclear Research Centre, SCK-CEN, Radioecology Section, Radiation Protection Department,  
Boeretang 200, B-2400 Mol, Belgium*

Indian mustard

Ryegrass



# Bioavailability Is essential

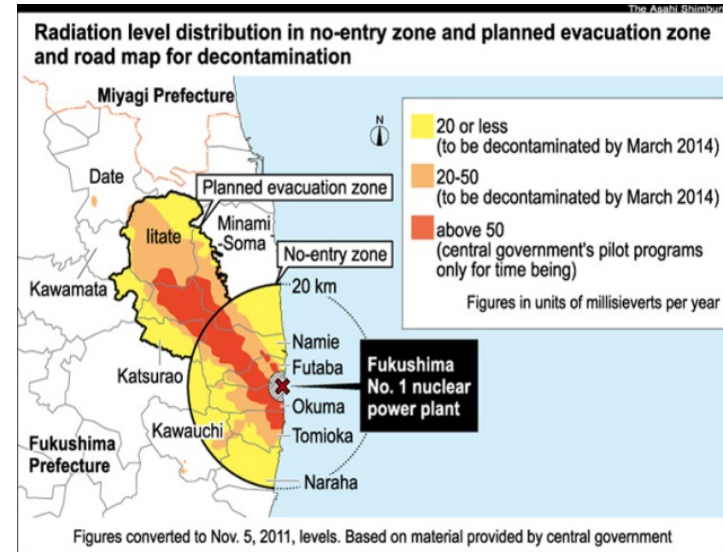
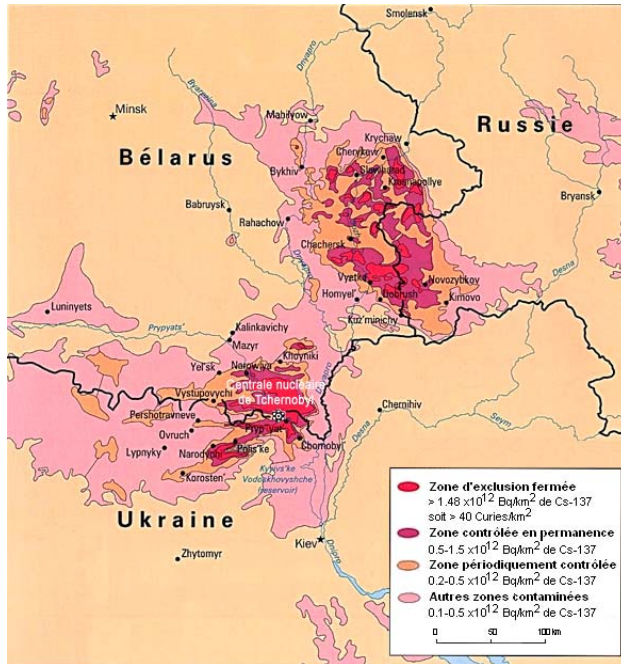


Uptake of <sup>239</sup>Pu by Indian mustard (*Brassica juncea*) and sunflower (*Helianthus annuus*) from different soils as affected by Pu-nitrate with DTPA applications

Pu source	Activity level of Pu (Bq g <sup>-1</sup> )	Amounts of DTPA applied	Pu uptake <sup>a</sup>			
			Crowley soil (Bq g <sup>-1</sup> )		Weswood soil (Bq g <sup>-1</sup> )	
			Indian mustard	Sunflower	Indian mustard	Sunflower
<sup>239</sup> Pu(NO <sub>3</sub> ) <sub>4</sub>	44.40	0	8.63 ± 1.92 a	2.47 ± 0.50 b	1.50 ± 0.40 b	1.77 ± 0.43 b
		10	208.69 ± 30.68 a	137.48 ± 6.38 b	90.95 ± 10.61 c	15.02 ± 2.66 d
		50	247.11 ± 33.13 a	269.15 ± 33.53 ab	163.93 ± 8.26 c	41.38 ± 7.61 d
<sup>239</sup> Pu(NO <sub>3</sub> ) <sub>4</sub>	222.0	0	25.88 ± 3.59 a	4.09 ± 0.49 b	3.84 ± 0.38 b	1.84 ± 0.45 b
		10	338.77 ± 16.28 a	168.98 ± 11.42 b	273.57 ± 19.66 c	65.82 ± 4.64 d
		50	788.48 ± 104.55 a	907.83 ± 47.67 b	349.85 ± 64.27 c	112.17 ± 7.80 d

With DTPA, increase in uptake by 30 to 100 fold

# Cs is the most extended contaminant



## Plants recycle soil Cs

## Cs moves slowly in soils

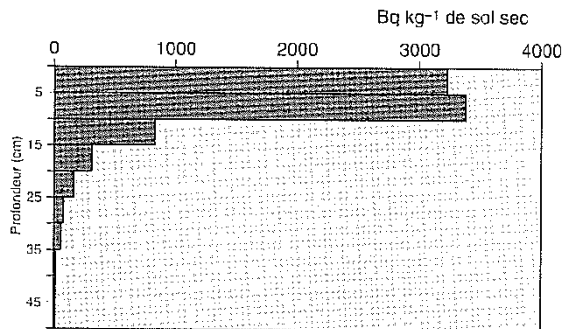
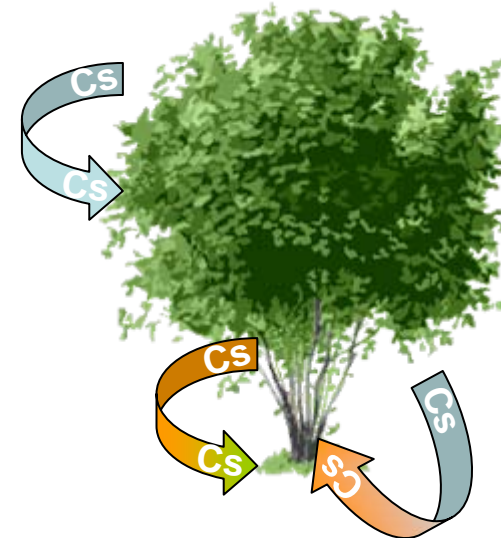
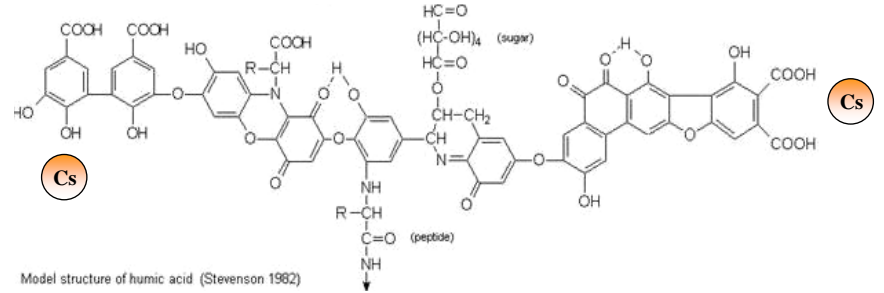


Figure 5.3. Répartition en profondeur du <sup>137</sup>Cs dans un sol de vigne 30 ans après son apport dans les 10 premiers centimètres de la couche de surface (Grauby, 1993).

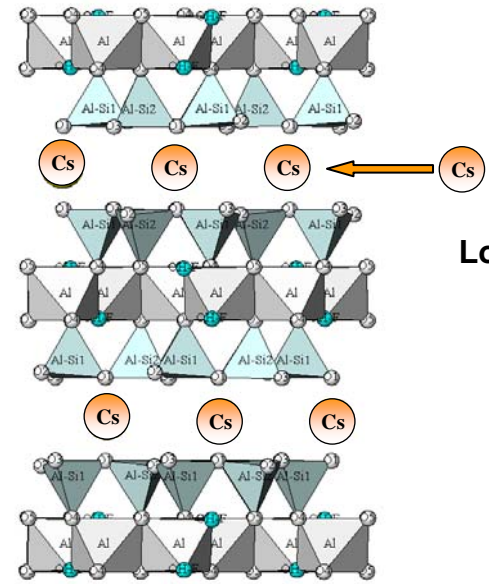
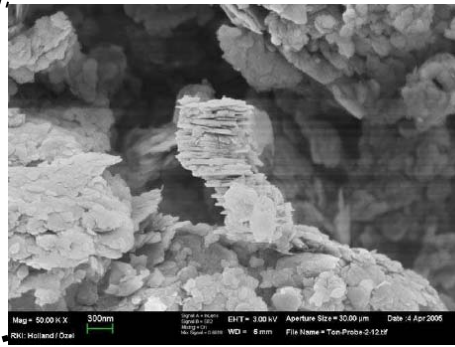


# Cs interacts with soil particles

exchangeable



Strongly fixed





## DIFFERENCES IN ROOT UPTAKE OF RADIOCAESIUM BY 30 PLANT TAXA

Martin R. Broadley and Neil J. Willey\*



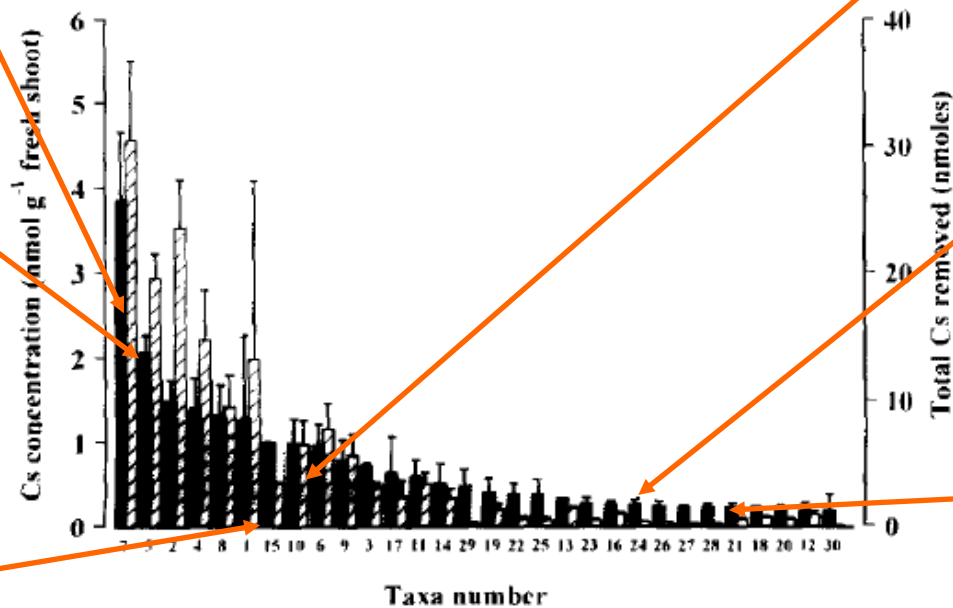
quinoa



sugar beet



atriplex



ray-grass

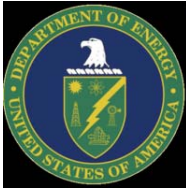


Festuca



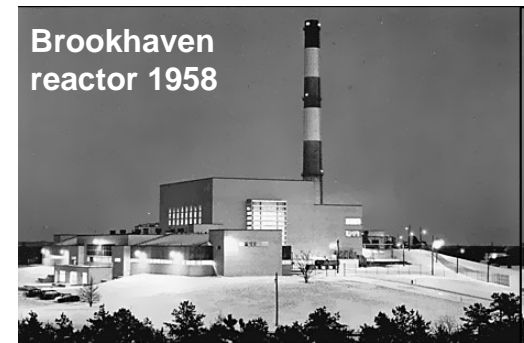
clover

**Fig. 1.** Shoot Cs concentration and total Cs removed after 6 h exposure to 20 ml of radiolabelled 10  $\mu$ mol CsCl (Error bars represent standard error of the mean; for taxa numbering, see Table 1).



# Cs phytoextraction the Brookhaven example

kBq/kg  $^{90}\text{Sr}$  0.02 – 1.4,  $^{137}\text{Cs}$  0.02-110



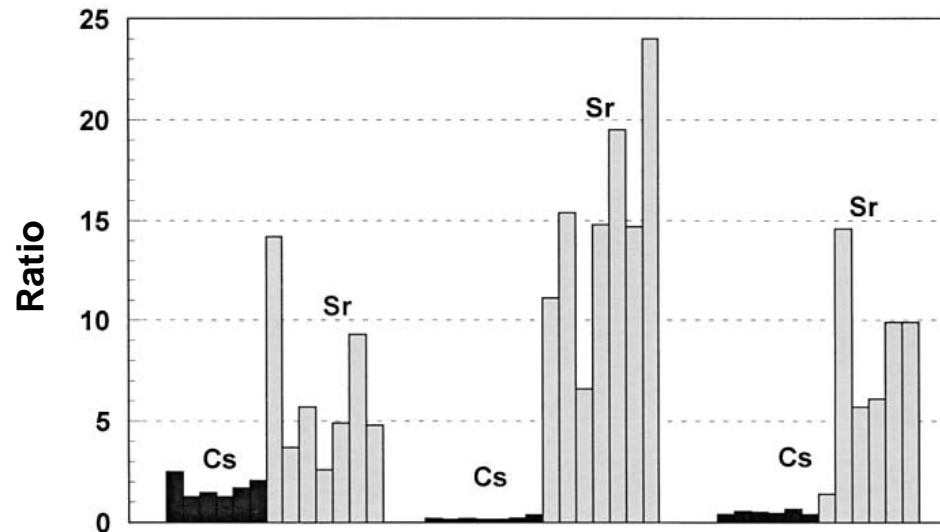
*Amaranthus retroflexus*



*Brassica juncea*



*Phaseolus acutifolius*



Fuhrmann et al. *J Environ Qual* 2002

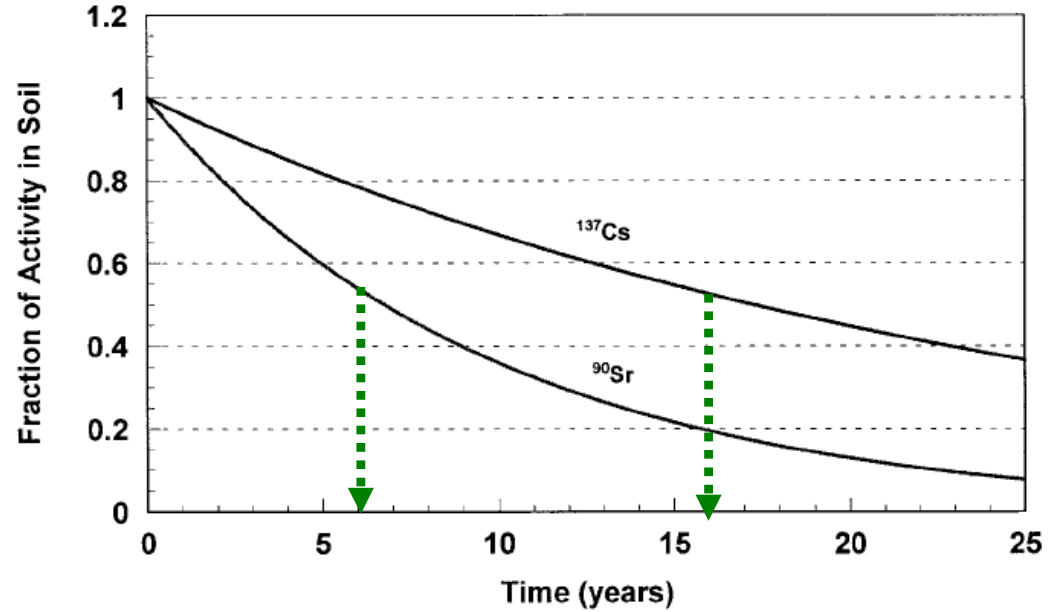


# Cs phytoextraction; the Brookhaven example

kBq/kg      $^{90}\text{Sr}$  0.02 – 1.4,  $^{137}\text{Cs}$  0.02-110



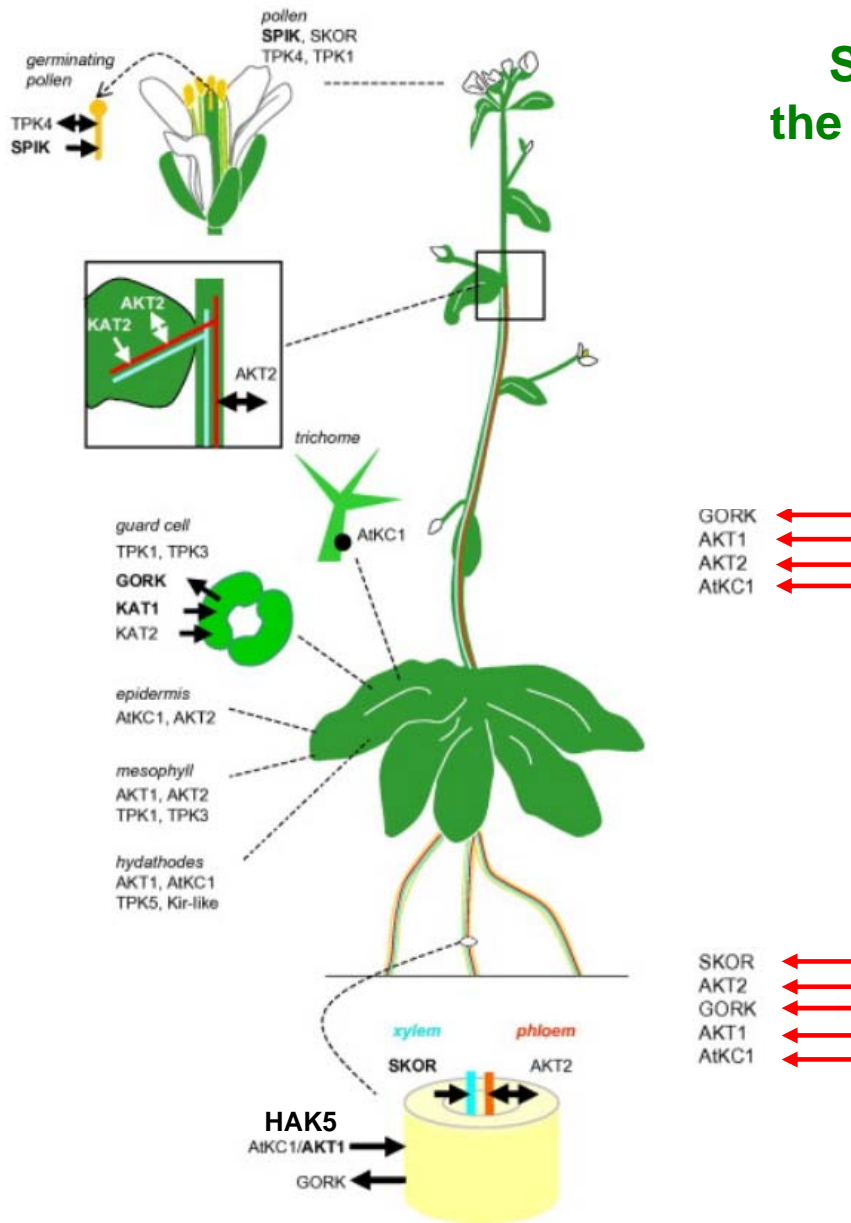
*Amaranthus retroflexus*



## Soil plant transfer of Cs What are the molecular processes ?

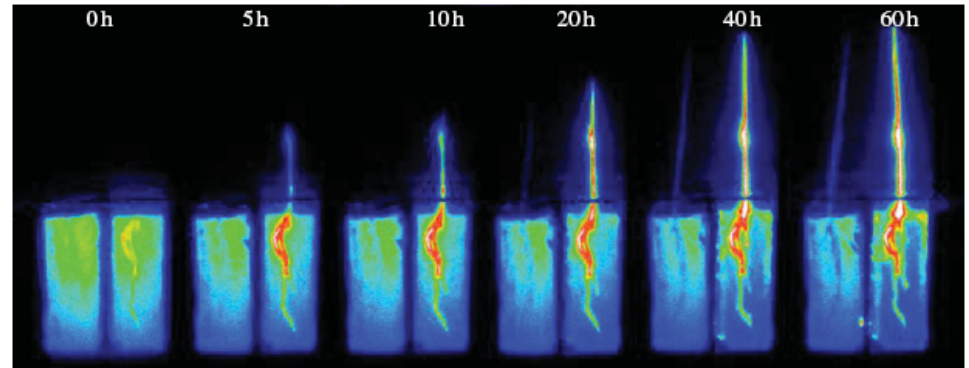
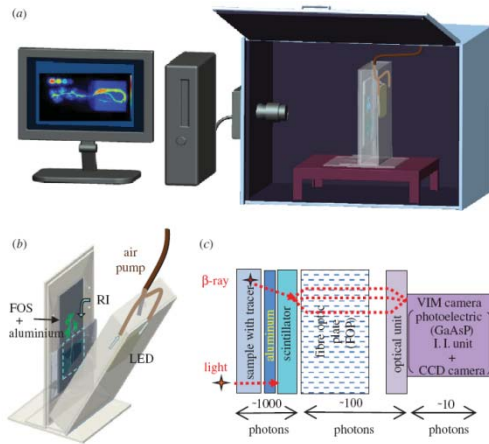


# Screening genes to understand the processes of Cs uptake in plants



Arabidopsis

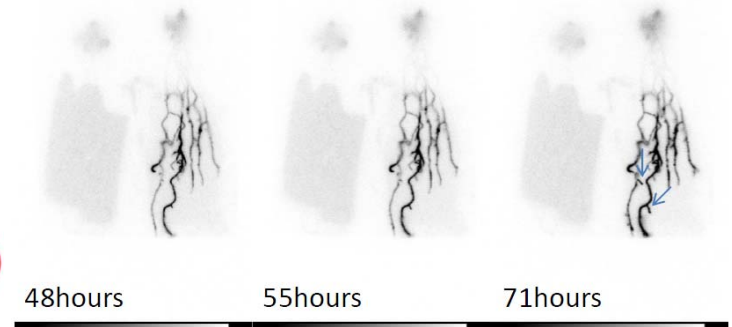
# Screening genes to understand the processes of Cs uptake in plants

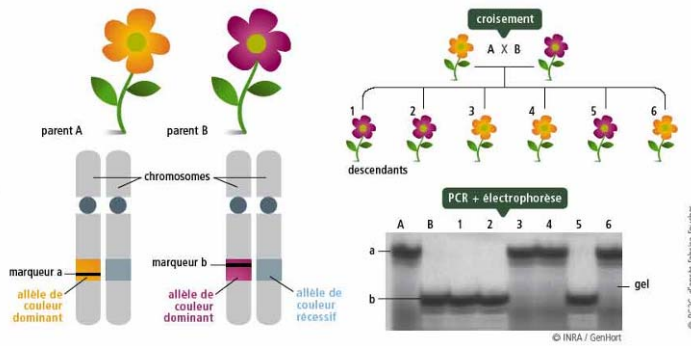


-K      -K

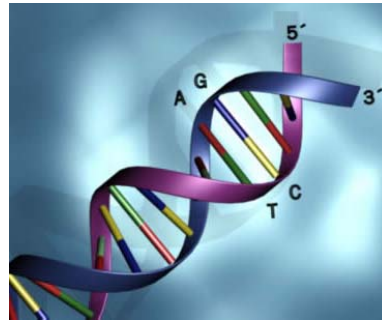
K5 $\mu$ M      K0 $\mu$ M  
Cs1 $\mu$ M      Cs1 $\mu$ M

+ 137Cs      + 137Cs  
50kBq(2.7 $\mu$ Ci)      50kBq(2.7 $\mu$ Ci)

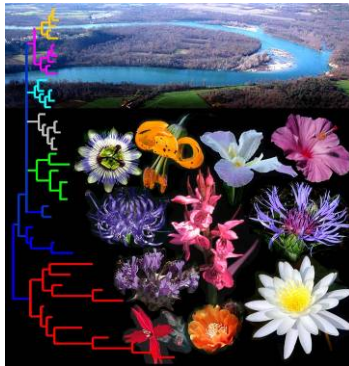




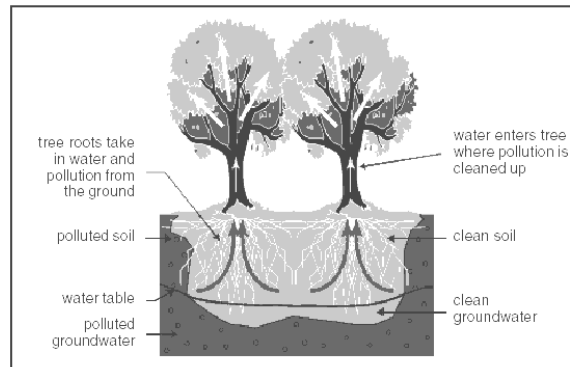
## Genetics



## Identification of molecular markers



## Biodiversity

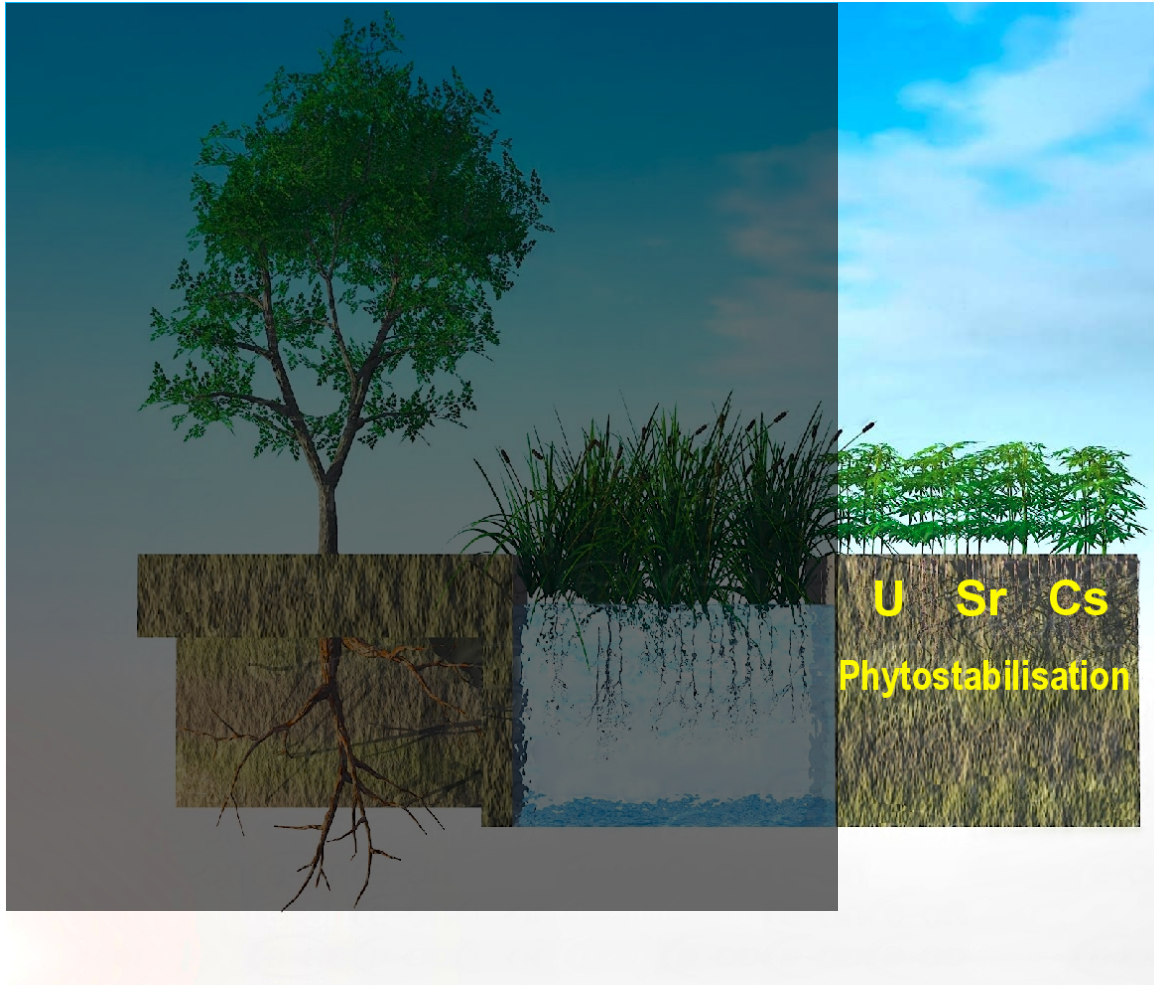


## Biotechnologies

## Safe food

## Phytoremediation

# Phytostabilization





# Phytostabilisation of arsenic

La Combe du Saut, projet DIFPOLMINE

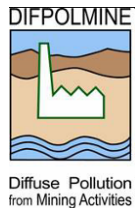


Figure 33. La Combe du Saut in December 2006.

## Phytostabilization

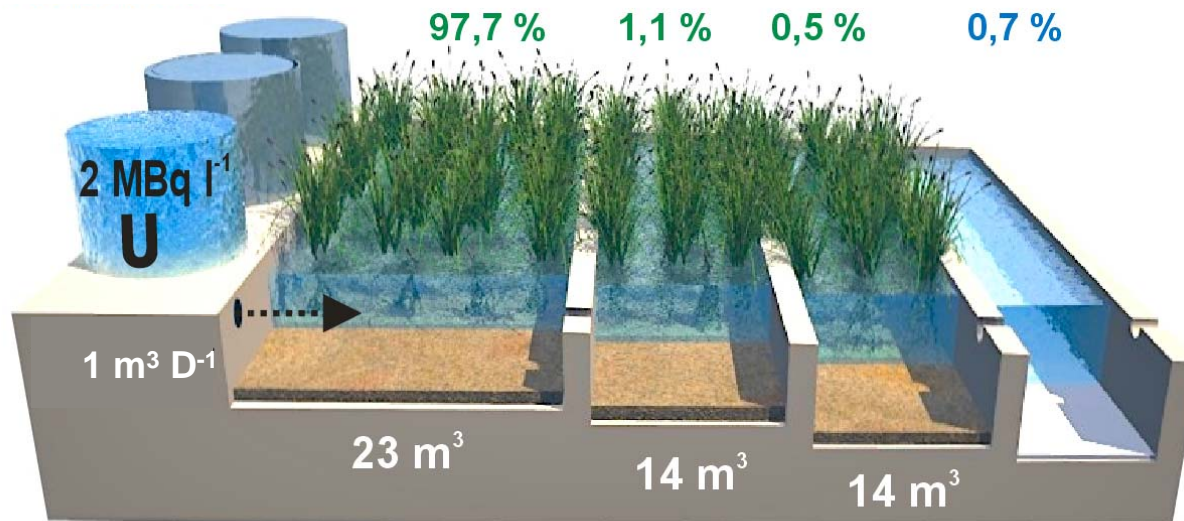
# Rhizofiltration



# Phytoremediation of Uranium by successive ponds

*Ex situ*

Rhizofixation



Timofeeva-Ressovskaia. *Proc. Inst Biol.* 1963

# Phytoremediation of Uranium in Tchernobyl and Ashtabula

Ashtabula (OHIO) (U, Tc, TCE)



Tchernobyl (U)



Injection of polylactate ester, acetate



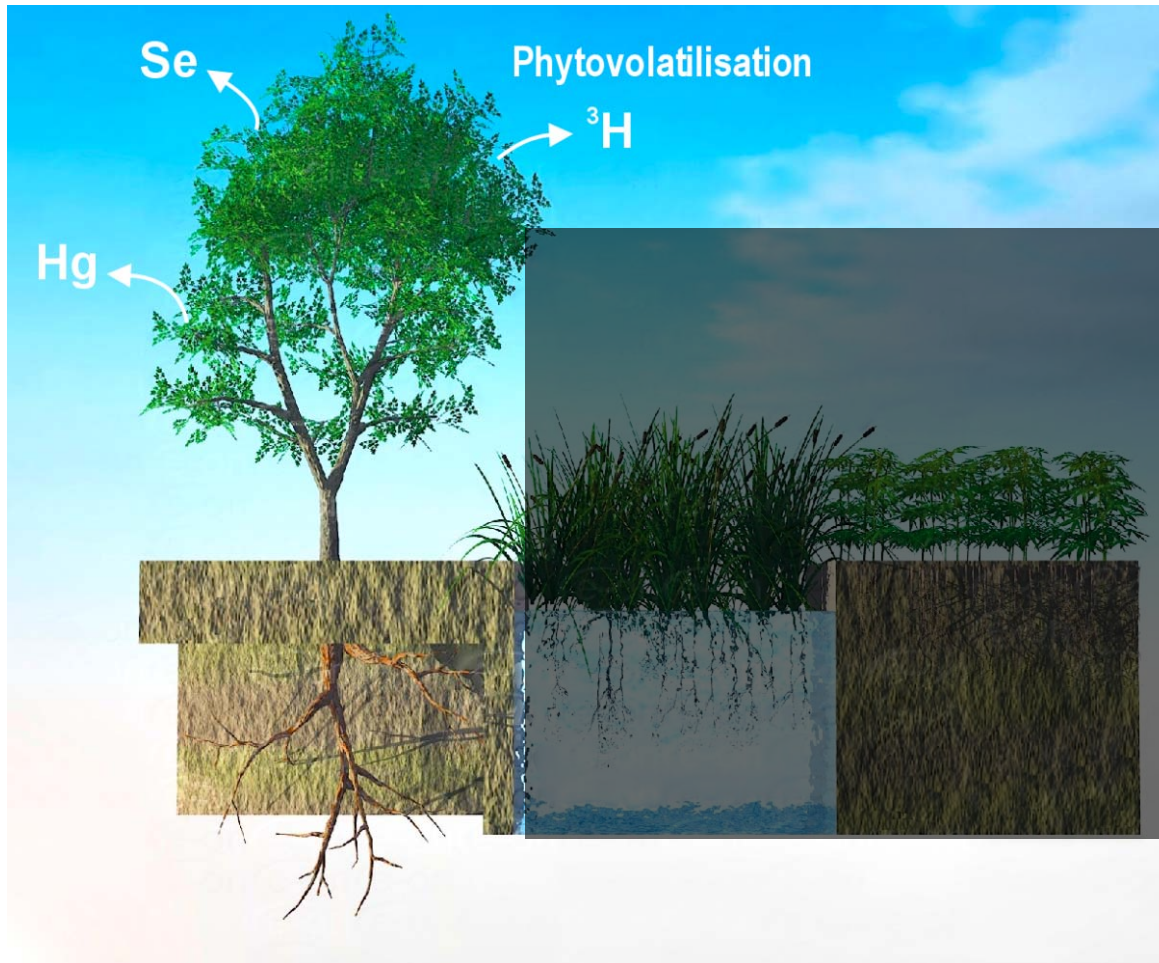
*Geobacter* U(VI) → U(IV)



rhizofiltration *In situ*

TF 5000-30000 in roots at pH 5,5

# Phytovolatilization



# Phytovolatilization



Enrico Fermi, December 1942  
First controlled chain reaction



United States  
Environmental Protection  
Agency

Region 5  
Superfund Division

EPA-905-B-04-001  
August 2004  
[www.epa.gov/region5superfund](http://www.epa.gov/region5superfund)  
[clu-in.org](http://clu-in.org)

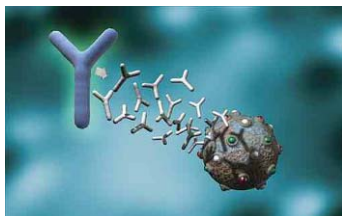
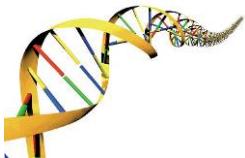


# Radionuclide Biological Remediation Resource Guide



# Limits of bioremediation

- Duration of the treatment needs to be improved to reach a result in less than a decade
- Contamination must be moderate
- Each case needs a new scenario, a multiparametric system
- Highly dependent on the environment
- Exploitation of biomass increases the economic attractiveness of the technique
- Ideally must be combined with a cogenerating energy technology
- Needs further researches to improve efficiency
- Poorly attractive to private industry due to a stochastic demand





# Stakes and Limits of Bioremediation

## Thanks for your attention !

