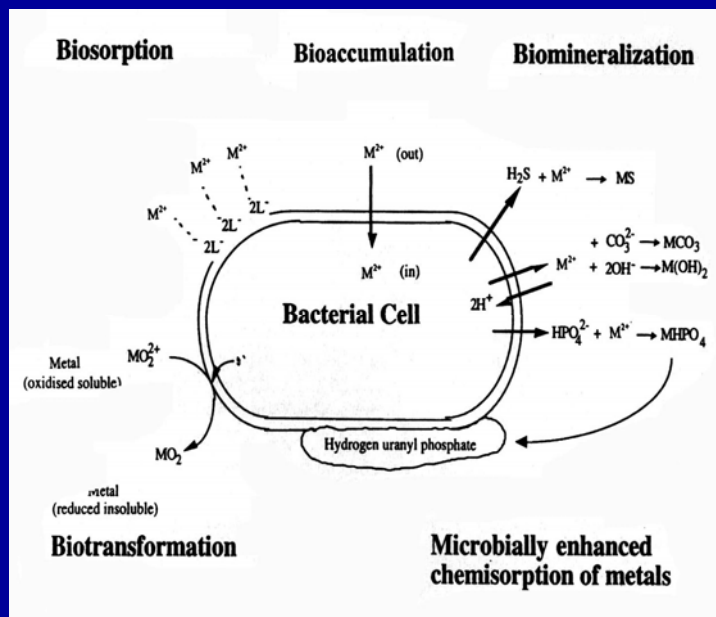


# Interaction of long-lived radionuclides and microorganisms

Khijniak T.V.

Winogradsky Institute of Microbiology of RAS

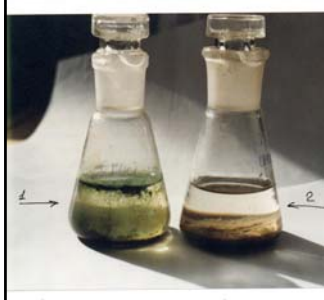
## Principal types of interaction



## Interaction of metals and microorganisms

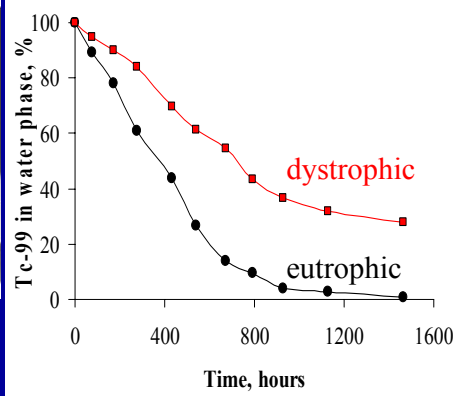
Type of biomass	Mechanism
Living cells Cell suspension or extract	1. Adsorption or complexation on cell wall 2. Intracellular accumulation 3. Oxidation or reduction of metals 4. Transformation: methylation or demethylation 5. Inorganic ligand formation and metal precipitation 6. Metal binding with the exopolymers
Dead biomass	1. Adsorption or complexation on cell wall 2. Metal precipitation under enzyme action
Microbial products	1. Siderophores, thioneines 2. Polimers of cell wall – bacterial peptidoglican, chitin or chitosan from fungi 3. Melanins, humic acids 4. Pigments – quinone

## Uptake of long-lived radionuclides by lake sediments of different trophic types



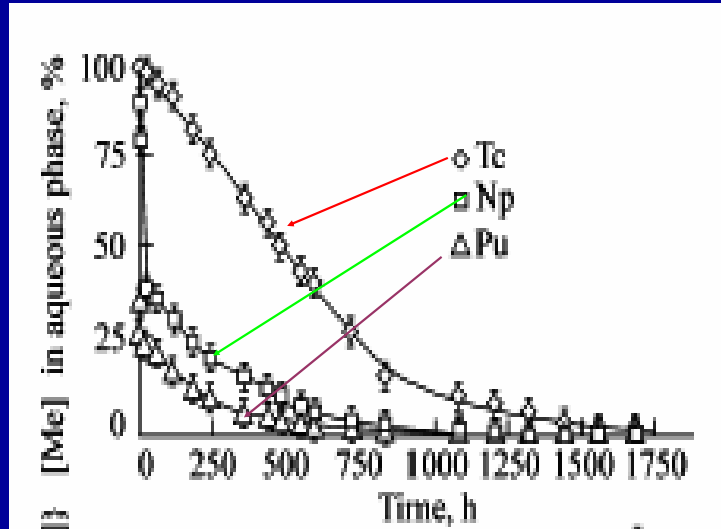
Solid phase –  
0,023 mg/ml

1 – 4 months experiment.  
2 – sterile controle (formalin or  $\gamma$ -irradiation)



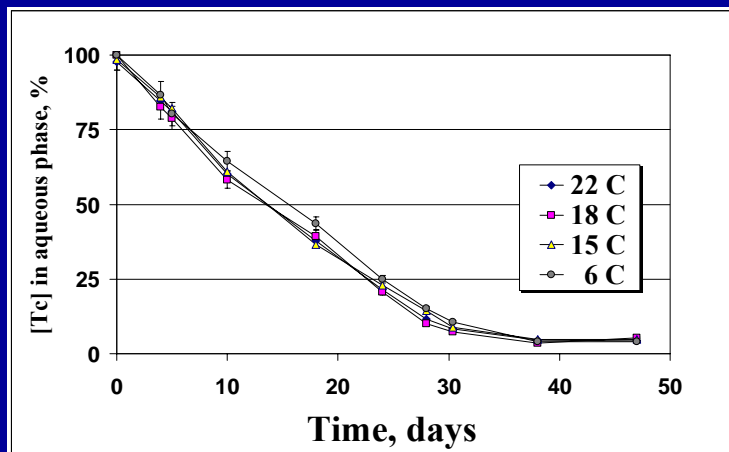
$t=15^{\circ}\text{C}$ ,  
 $[\text{Tc}]_0=10^{-4}\text{M}$

## Uptake of radionuclides by eutrophic sediments (lake Beloe, Moscow region)

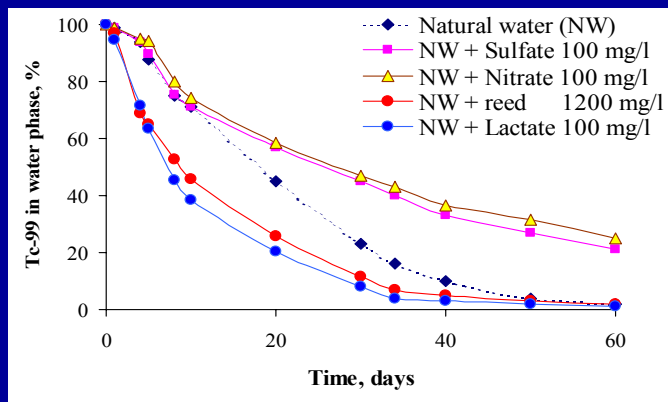


$$[Tc]_0 = [Np]_0 = [Pu]_0 = 10^{-5} \text{ M}$$

## Temperature influence on Tc-uptake by eutrophic sediment



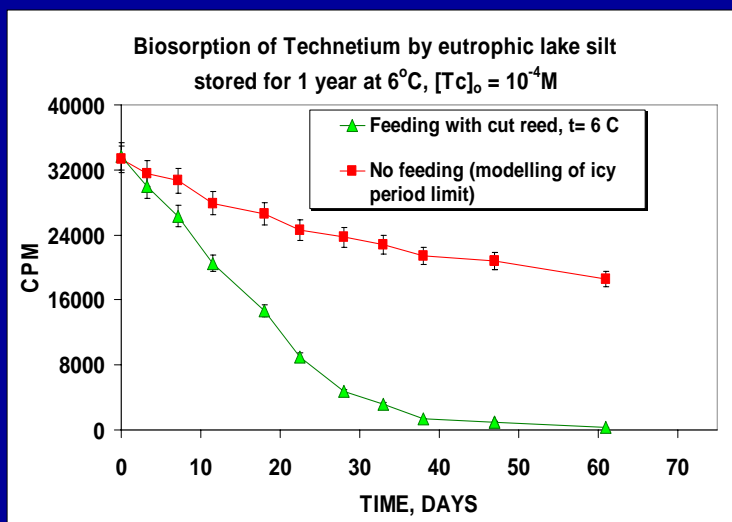
## Substrate impact on the $^{99}\text{Tc}$ uptake by eutrophic sediment



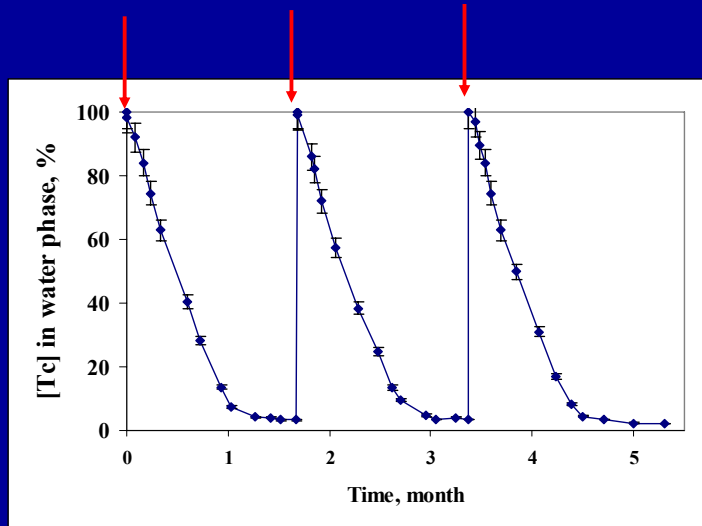
### Uptake rate of $^{99}\text{Tc}$

- \* Edition of e-acceptor (sulfate, nitrate) ↓
- \* Edition of e-donor (lactate, reed) ↑

## Substrate impact on the $^{99}\text{Tc}$ uptake by eutrophic sediment (2)

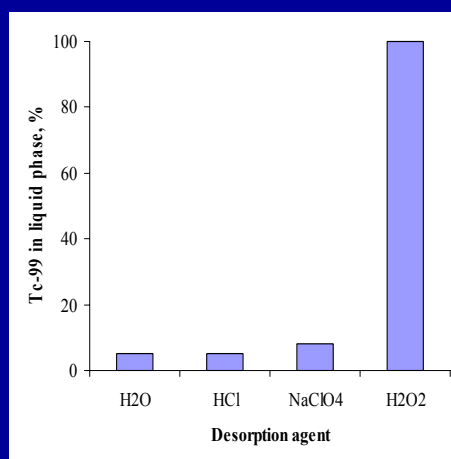


## Multiple addition and uptake of Tc-99 by eutrophic sediments



$t = 18^{\circ}\text{C}$ ,  $[\text{Tc}]_0 = 10^{-4}\text{M}$

## Desorption of $^{99}\text{Tc}$ from eutrophic sediments



Desorption factor:

- ❖ H<sub>2</sub>O - 0.05
- ❖ HCl - 0.05
- ❖ NaClO<sub>4</sub> - 0.08
- ❖ H<sub>2</sub>O<sub>2</sub> - 0.99

## Tc-reduction by microorganisms



## Bacterial reduction of $\text{TcO}_4^-$

- ❖ *Desulfovibrio desulfuricans*
  - ❖ *Desulfovibrio gigas*
  - ❖ *Moraxella*, *Planococcus*
  - ❖ *Shewanella putrificiens*
  - ❖ *Geobacter metallireducens*
  - ❖ *Escherichia coli*
  - ❖ *Pseudomonas vanadiumreductans*
  - ❖ *P. isachenkovii*
  - ❖ *Thermoterrabacterium ferrireducens*
  - ❖ *Tepidibacter thalassicus*
  - ❖ *Pyrobaculum islandicum*
- } pH 7
- ❖ *Acidithiobacillus ferrooxidans*
  - ❖ *Acidithiobacillus thiooxidans*
- } pH 2-3
- ❖ *Halomonas sp.*
- pH 9-10

## Reduction of $^{99}\text{Tc O}_4^-$ by suspension of sulfate-reducing bacteria

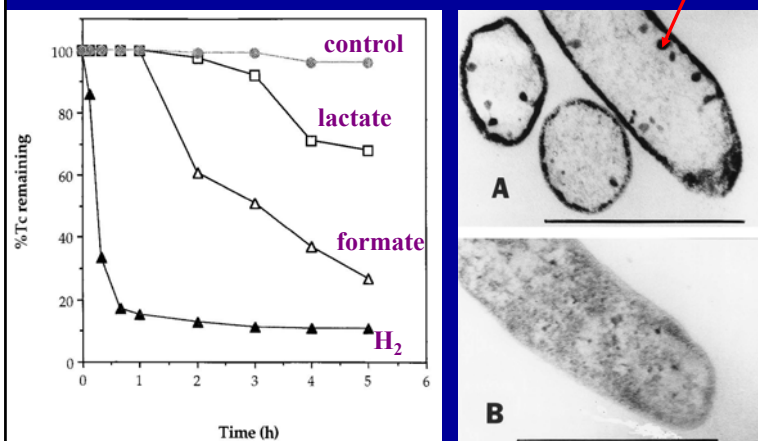
TABLE 1. Effect of electron donor on Tc reduction and removal by resting cells of *D. desulfuricans*<sup>a</sup>

Electron donor	CR
Hydrogen .....	12,850
Formate.....	11,648
Pyruvate.....	6,393
Lactate.....	2,940
Ethanol.....	1,093
Succinate .....	363
Glycerol.....	336
Acetate .....	197
Methanol.....	101

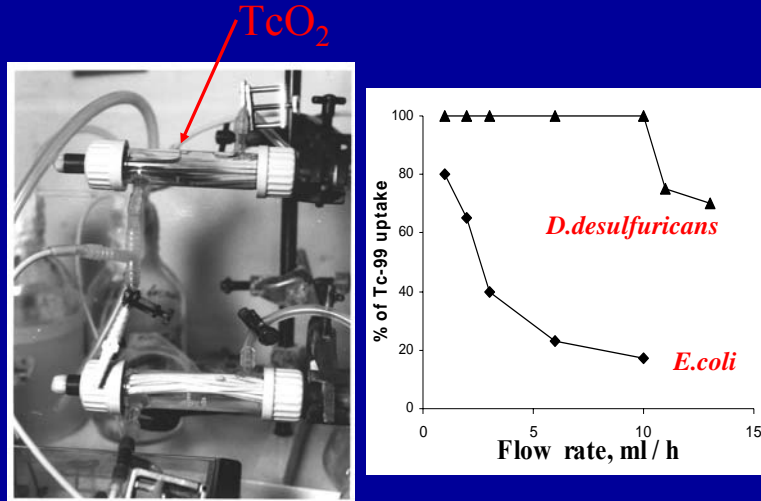
<sup>a</sup> Results are expressed as the CR, calculated after 24 h of incubation at 30°C.

*Desulfovibrio desulfuricans*

## Kinetic of $^{99}\text{Tc O}_4^-$ reduction by *D. desulfuricans*

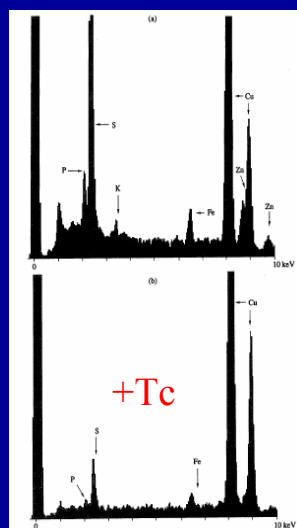


## Reduction of $^{99}\text{TcO}_4^-$ by sulfate-reducing bacteria



pH=7,  $[\text{TcO}_4^-]_0 = 25 \text{ mg/L}$   
*Desulfovibrio desulfuricans*

## Impact of pertechnetate on metabolism of acidithiobacilli



*Acidithiobacillus ferrooxidans*

Technetium concentration, mg/l	Concentration of $\text{Fe}^{3+}$ formed, mg/l	Protein concentration at the end of the experiment, $\mu\text{g/ml}$
0	2500–2800	75
50	275–285	35
100	200; 400	30; 45
250	50–60	15
500	0	10
750	0	10

pH 2.5



## Reduction of $\text{TcO}_4^-$ under anaerobic condition, pH 2.5, incubation 7 d

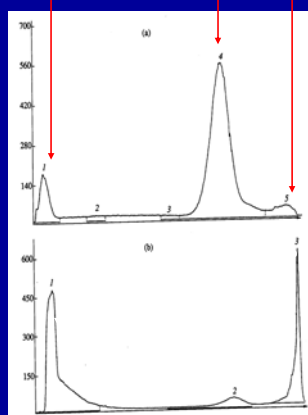


*Acidithiobacillus ferrooxidans*

№	Final radioactivity of solution, %
Control	100
1	43
2	39

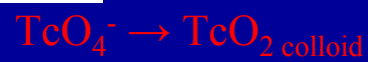
## Chromatographic separation of Tc-99 after incubation with *A.ferrooxidans*

$\text{Tc}^{5+}$        $\text{Tc}^{7+}$     $\text{Tc}^{4+}$       pH 2.5



Bacterial spent medium

Colloid



## Reduction of $\text{TcO}_4^-$ by haloalkaliphilic bacteria genus *Halomonas*

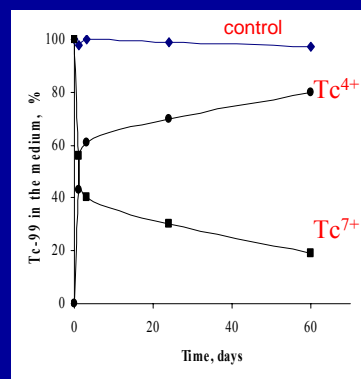
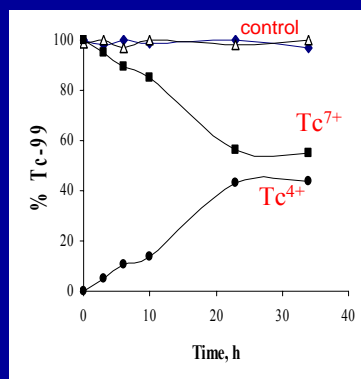


pH = 10,  
 $[\text{Tc}]_0 = 0.25 \text{ mM}$

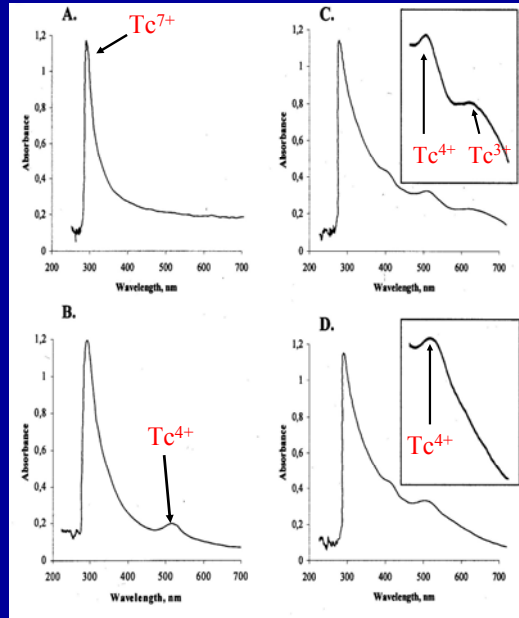
1 - control  
 2,3 - *Halomonas*

## Kinetic of bacterial reduction of $\text{TcO}_4^-$ under alkaline conditions

pH 9,8-10, carbonate/bicarbonate medium, salinity 1 M,  
*Halomonas* strain Mono, acetate 10 mM,  $\text{TcO}_4^- = 0.25\text{mM}$

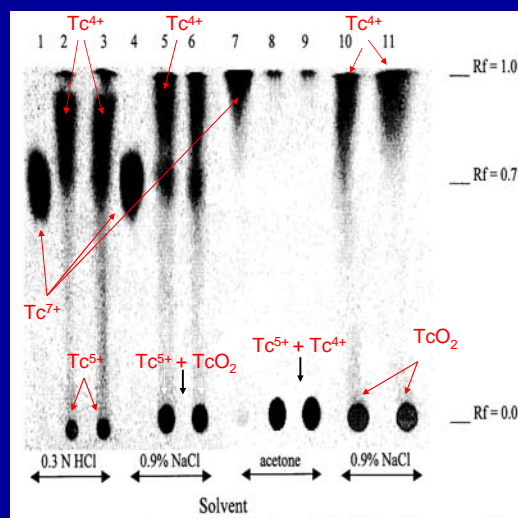


## Spectrophotometric analysis of bacterial spent medium under alkaline conditions



Anaerobic conditions  
pH 9,8-10,  
salinity 1 M,  
*Halomonas* strain Mono  
Acetate 10 mM,  
 $\text{TcO}_4^- = 0.25 \text{ mM}$

## Chromatographic separation of different forms of Tc



$\text{TcO}(\text{OH})_2 (\text{aq})$   
 $\text{TcO}(\text{OH})_3^-$   
 $\text{Tc}(\text{OH})_2\text{CO}_3$   
 $\text{Tc}^{\text{IV}}\text{O}(\text{OH})_3(\text{CO}_3)^-$

Anaerobic conditions, pH 9,8-10, salinity 1 M,  
*Halomonas* strain Mono, Acetate 10 mM,  $\text{TcO}_4^- = 0.25 \text{ mM}$

## Influence of e-donors on pertechnetate reduction under alkaline conditions

Substrate	Technetium in the supernatant (%)		
	Tc(VII)	Tc(IV)	Tc(V)
Formate	65.0	32	3
Acetate	67.0	28.9	4
Lactate	68.0	28	3
Methanol	69.0	29	2
Ethanol	68.8	28.8	2.4
Control without bacteria	100.0	0	0
Control without e-donor	100.0	0	0

Anaerobic conditions, pH 9,8-10, e-donors – 10 mM  
 salinity 1 M, suspension of strain Mono (0,035 mg/mL),  $\text{TcO}_4^- = 0.25\text{mM}$

## Conclusion – bacterial reduction of $\text{TcO}_4^-$

	Final reduced form	
pH 2-3	$\text{TcO}_2$ –colloid	!!! SOLUBLE
pH 7	$\text{TcO}_2$	Solid
pH 8-11	$\text{Tc}^{\text{IV}}\text{O}(\text{OH})_3(\text{CO}_3)^-$	!!! SOLUBLE

## Author grateful to :

- Medvedeva-Lyalikova N.N.
- German K.E.
- Peretrukhin V.F.
- Lloyd J.R.
- Simonoff M.