

# Rehabilitation of heavy metal contaminated brownfields using woody plants and mycorrhizal symbiosis

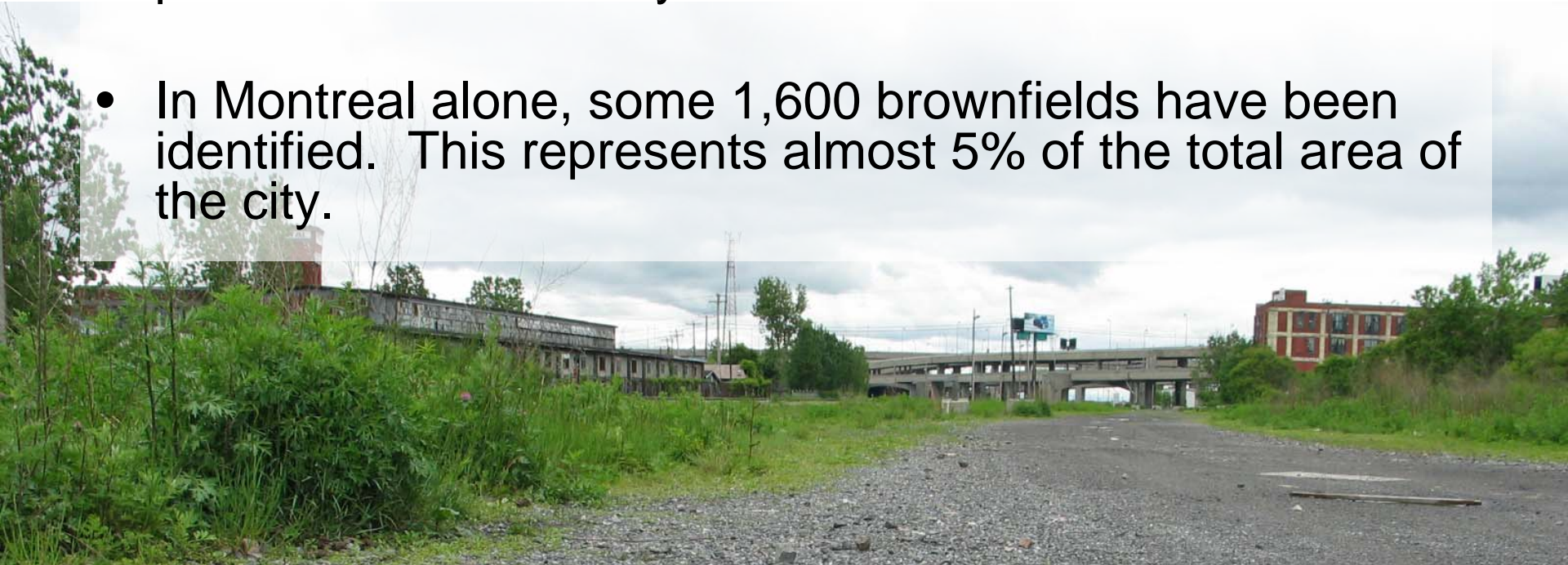
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# Context

- The National Round Table on the Environment and the Economy (NRTEE) reported that there are as many as 30,000 brownfield sites across Canada (old railway yards, former gasoline stations, etc.)
- These brownfields disfigure neighbourhoods and may pose health and safety risks.
- In Montreal alone, some 1,600 brownfields have been identified. This represents almost 5% of the total area of the city.





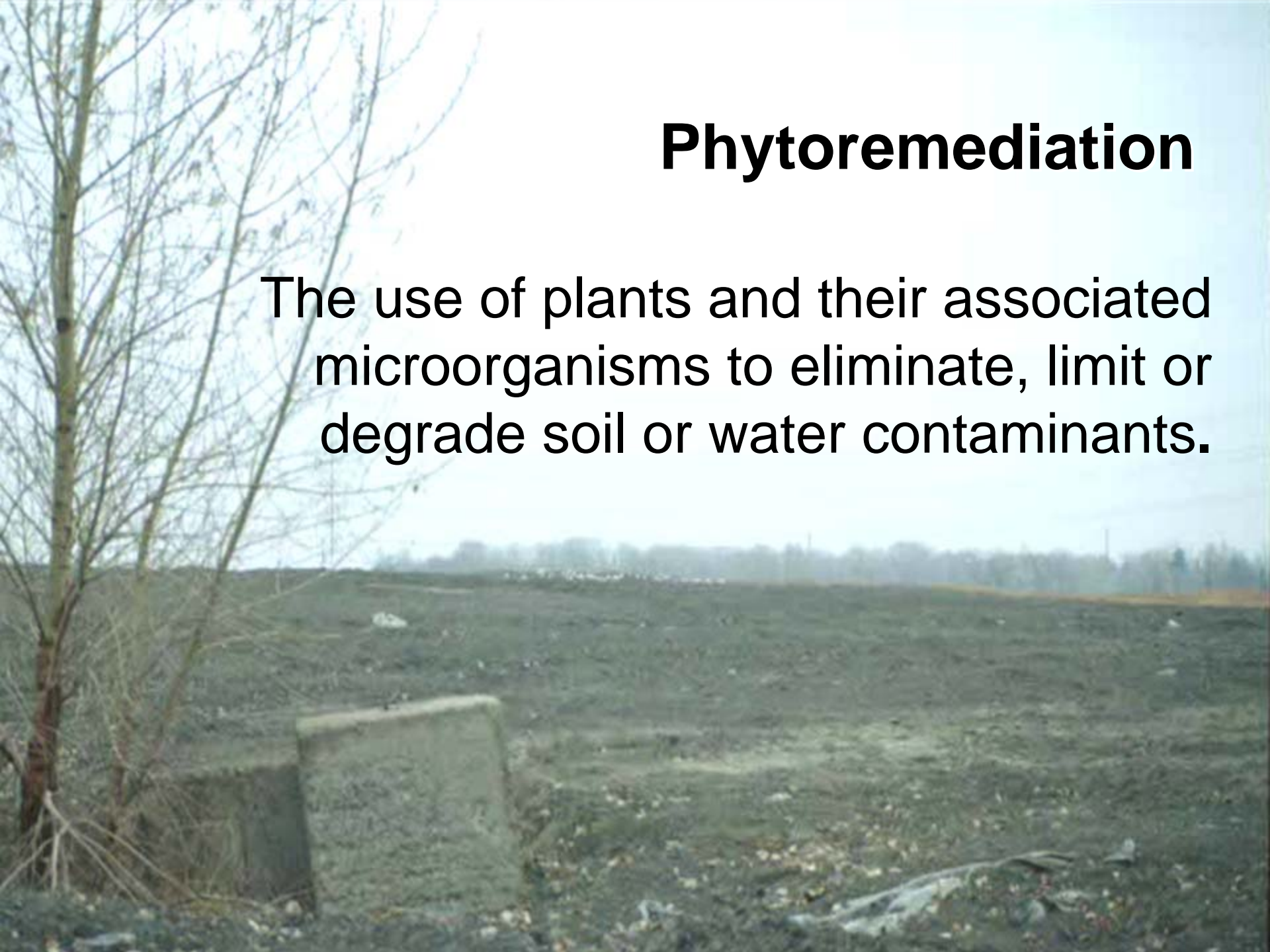
# Cleanup vs costs

- Cleanup costs are considered too expensive relative to income generated ( $> 1$  M dollars  $\text{ha}^{-1}$ ).
- Alternatives must be developed to provide the opportunity to treat these sites at lower cost.
- Green techniques using phytoremediation approaches can be an effective option.



# Phytoremediation

The use of plants and their associated microorganisms to eliminate, limit or degrade soil or water contaminants.





# Soil characteristics of urban brownfields

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- **Low level of organic matter content;**
- **Heavy and compacted soils;**
- **High pH;**
- **Poor drainage;**
- **Contamination of various origins (organic and inorganic);**
- **Very heterogeneous.**

# **Characteristics essential for plants used in phytoremediation of heavy metal contaminated brownfields**

- **Facility of establishment;**
- **High yield in climatic and edaphic conditions of urban areas;**
- **Good root development capacity;**
- **Capacity to absorb large quantities of metals.**









# Potential of hyperaccumulator species for phytoremediation

- ***Armeria maritima***  
(Plumbaginaceae)
- ***Brassica juncea***  
(Brassicaceae)
- ***Festuca arundinacea***  
(Poaceae)
- ***Minuartia verna***  
(Caryophyllaceae)
- ***Thlaspi caerulescens***  
(Brassicaceae)
- ***Vernonia petersii***  
(Asteraceae)





# General observations

- **Difficult to establish in typical brownfield soil conditions.**
- **Hyperaccumulators tested generally had poor biomass production.**
- **Their low aboveground yield made them difficult to harvest and to manage.**



# Comparing diverse plants species for their phytoremediation potential

- **Verify establishment and growth potential of willows and poplars on brownfields.**
- **Compare their metal accumulation with one of the highest-performing hyperaccumulator plant species.**
- **Verify the impact of chelating agent on growth parameters and metal accumulation.**







sites  
industriels

Terrain  
Pitt

Canal  
Lachine

# Comparison of the phytoextraction potential of willow species with Indian mustard

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## Methods

- Three blocs 38,5 m<sup>2</sup>
- Three species (two willow clones and *B. juncea*)
- Two treatments: with and without EDTA<sup>1</sup> (20 mM)

1. Disodium ethylenediamine tetraacetate dihydrate



# Species studied

- *Brassica juncea*
- *Salix viminalis* (5027)
- *Salix myabeana* (SX67)







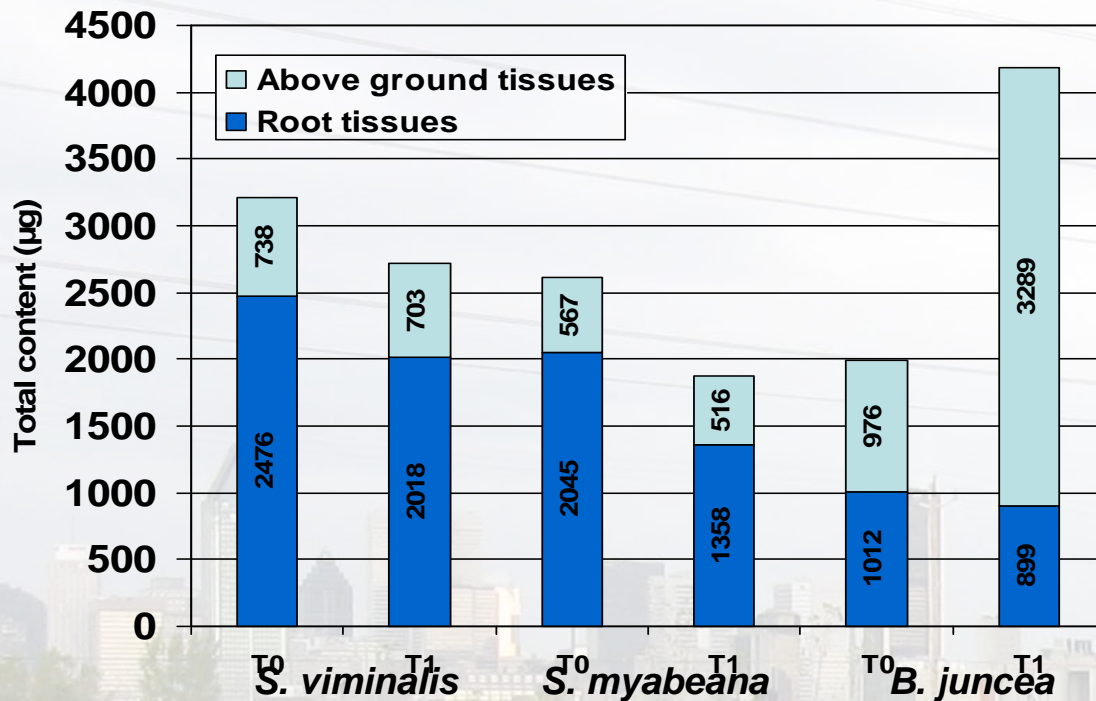


# Metal bioavailability

Metals	Exchangeable mg/kg <sup>-1</sup>	Carbonates mg/kg <sup>-1</sup>	Oxides mg/kg <sup>-1</sup>	Residues mg/kg <sup>-1</sup>	Total metals mg/kg <sup>-1</sup>	Criteria	Potential plant bioavailability mg/kg-1
Cu	x	38,4	96,2	282,2	416,8	B-C	38.4 to 134.6
Pb	x	162	311	384	857	B-C	162 to 473
Zn	0,6	149,4	419,6	329,8	899,4	B	149.4 to 569.6



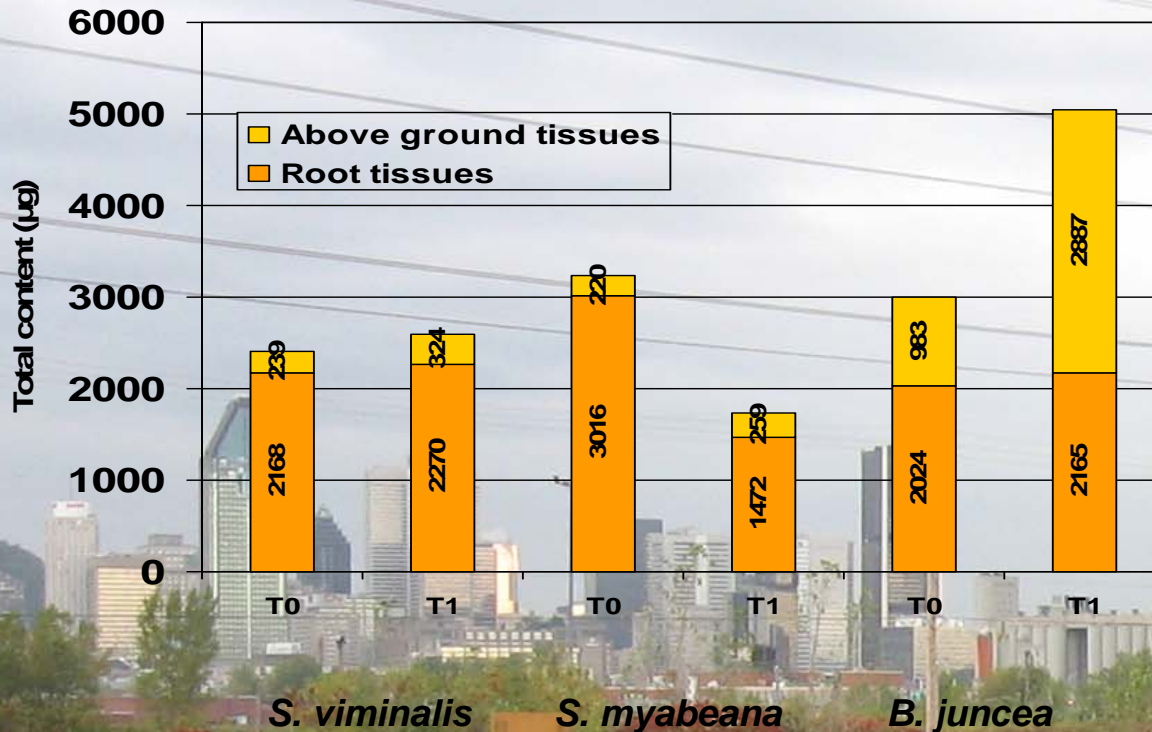
# Comparison of Cu content ( $\mu\text{g}$ ) in plants



- EDTA induced a significant effect on the accumulation of Cu in above ground tissues of *B. juncea*.
- EDTA had no impact on willows.
- Significantly more Cu in willow roots.

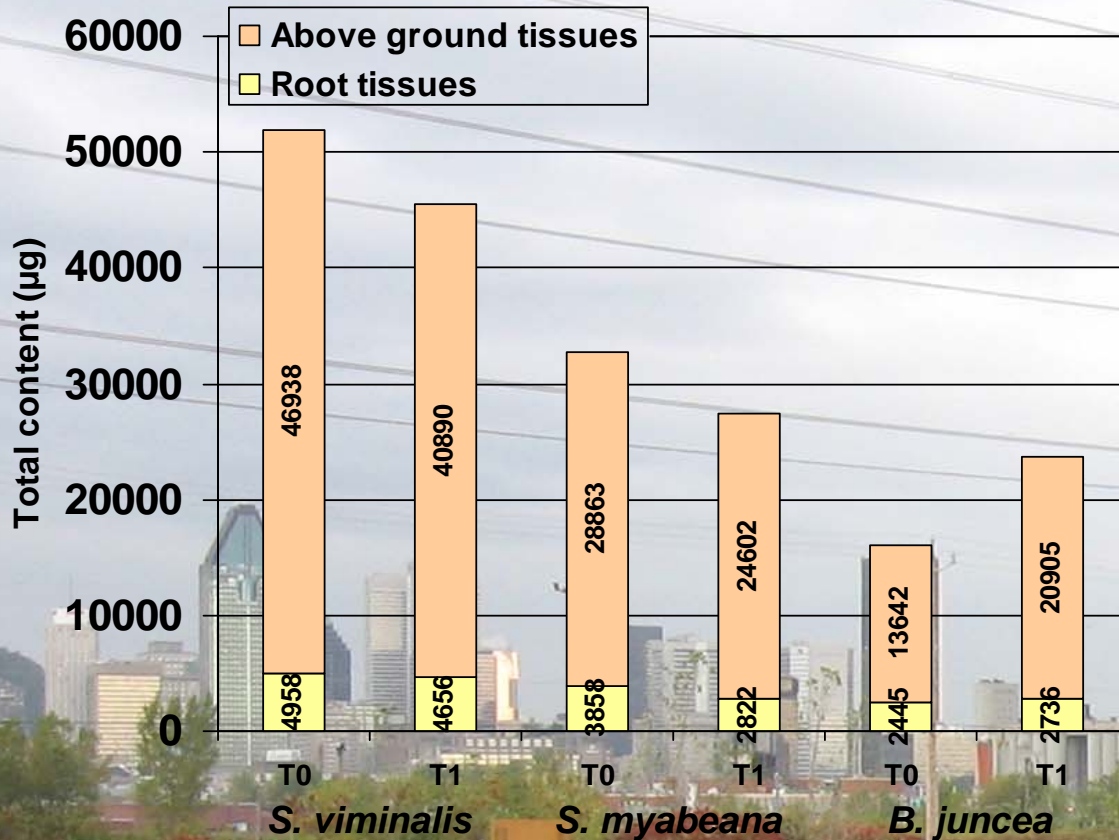


# Comparison of Pb content ( $\mu\text{g}$ ) in plants



- EDTA induced a significant effect on the accumulation of Pb in above ground tissues of *B. juncea*.
- EDTA had no impact on willows.
- Significantly more Pb in above ground tissues of *B. juncea*.
- No difference in roots of the three species.

# Comparison of Zn content ( $\mu\text{g}$ ) in plants



- EDTA had no effect either on above ground or root tissue Zn content in the three species.
- *S. viminalis* had significantly more Zn in above ground and root tissues.





# Conclusions

**Fast growing species such as willows are:**

- **Establish easily.**
- **Compete well in open fields.**
- **Efficiently absorb diverse contaminants and cumulate them in their tissues.**
- **Produce significant biomass, potentially with high contaminant content.**
- **Provide immediate visual impact (rapid growth).**
- **Facilitate harvest and treatment.**
- **Generate biomass that can be harvested and used for diverse applications.**

# A new experimental set up in 2006

## Objectives

- Compare both growth and capacity of two woody plant clones belonging to the genera *Salix* and *Populus* when inoculated or not with a commercial AM fungal inoculum containing *G. intraradices*, in heavy metal (Cd, Zn, Cu and Pb) contaminated brownfields;
- Evaluate the longer term HM phytoextraction potential of these clones.









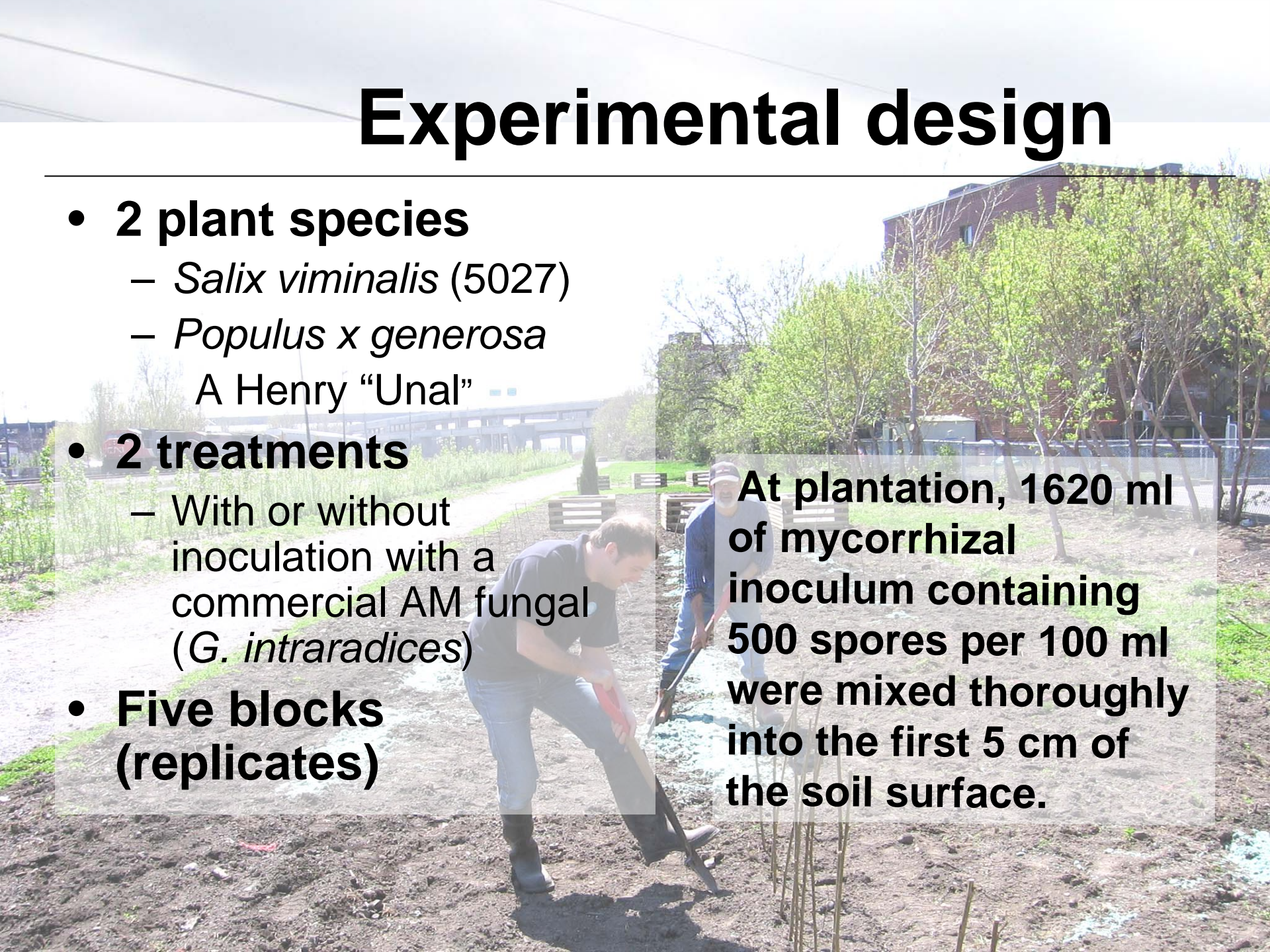


# Experimental design

- **2 plant species**
  - *Salix viminalis* (5027)
  - *Populus x generosa*  
A Henry “Unal”

- **2 treatments**
  - With or without inoculation with a commercial AM fungal (*G. intraradices*)

- **Five blocks (replicates)**



At plantation, 1620 ml of mycorrhizal inoculum containing 500 spores per 100 ml were mixed thoroughly into the first 5 cm of the soil surface.



















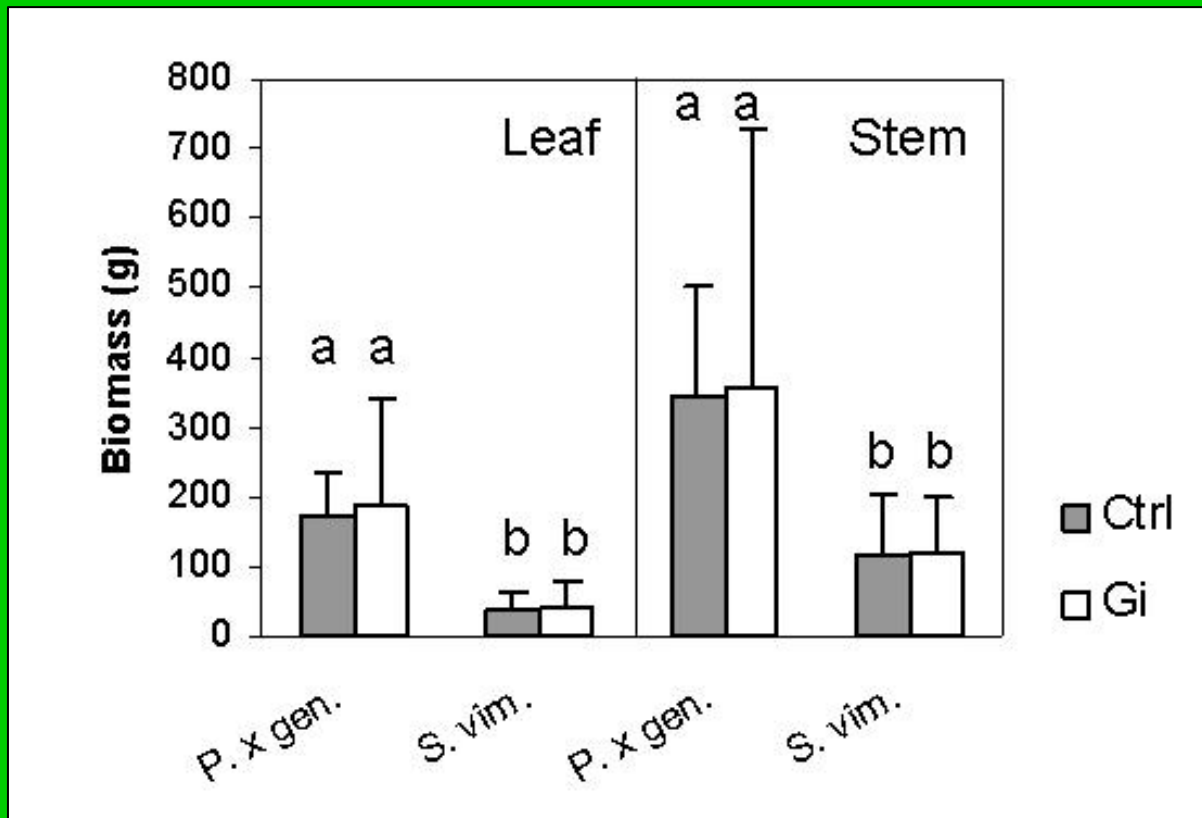
# Methods

**In September 2007 (at the end of the second growing season following establishment:**

- Samples of leaves, stems and roots were taken.**
- Zn, Cu, Cd and Pb concentrations were determined.**



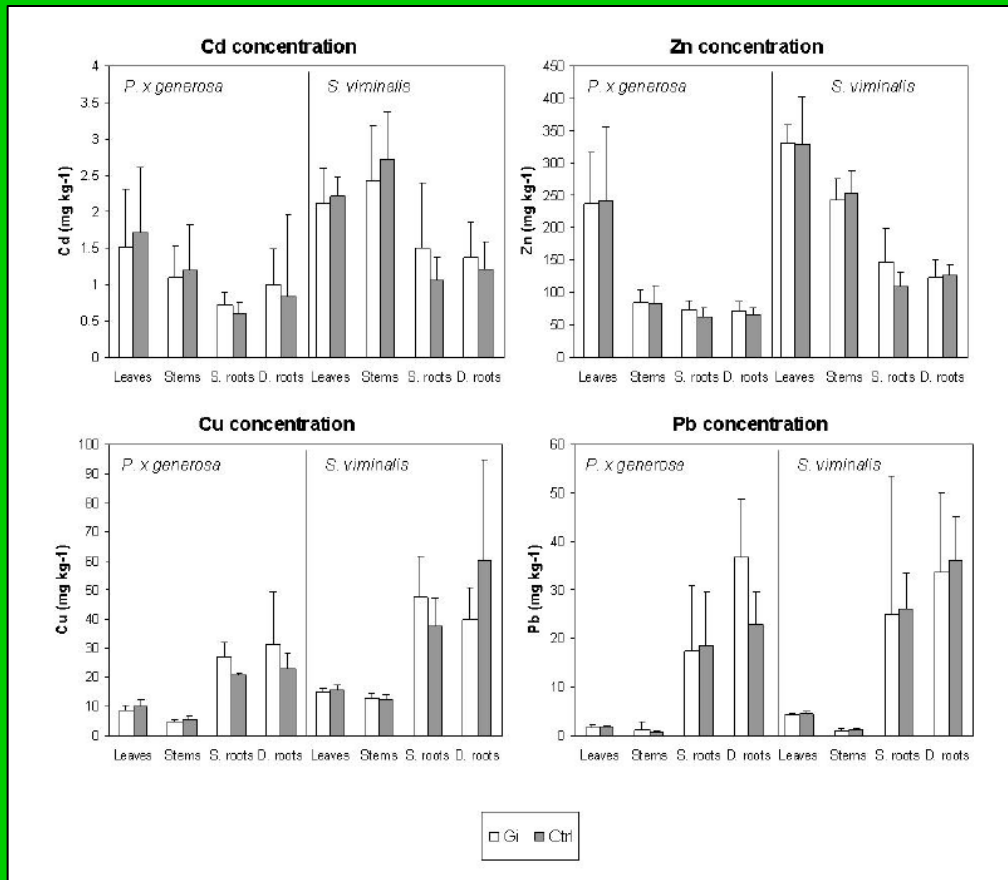
Dry leaf and stem biomass production per plant of *P. × generosa* and *S. viminalis* clones, non-inoculated (Ctrl) or inoculated (Gi) with *G. intraradices*, measured after two growing seasons.



N.B. Within each tissue, columns with a different letter are significantly different at  $p < 0.05$ ; there was no significant difference between inoculation treatments ( $n = 40$ ). Bars represent standard deviation.



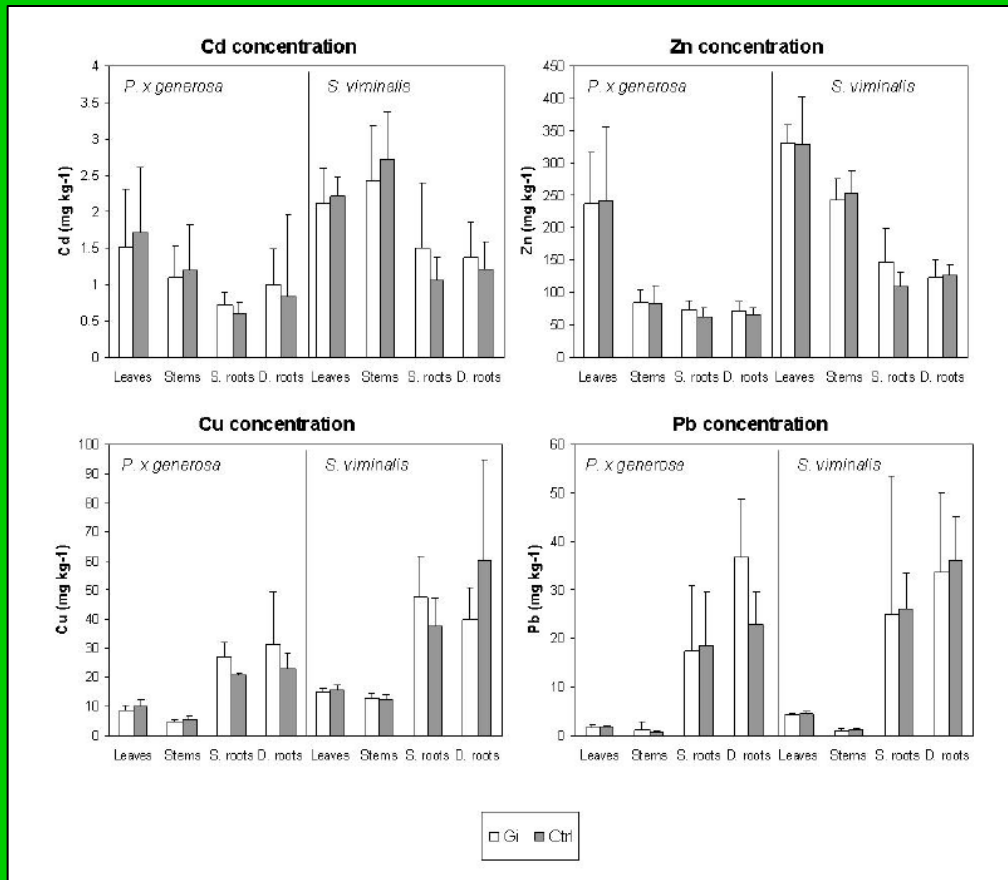
# Heavy metal concentrations measured in *P. × generosa* and *S. viminalis* tissues, inoculated (Gi) or non-inoculated (Ctrl) with *G. intraradices*, at the end of the second year of growth.



ANOVA comparisons <sup>a, b</sup>				
	Leaves	Stems	Surface roots	Deep roots
Cd	Sv=Pg	Sv>Pg***	Gi: Sv=Pg** Ctrl: Sv>Pg**	Sv>Pg**
Zn	Sv=Pg	Sv>Pg***	Sv>Pg*	Sv>Pg**
Cu	Sv>Pg**	Sv>Pg***	Sv>Pg*	Sv>Pg*
Pb	Sv>Pg***	Sv=Pg	Sv=Pg	Sv=Pg



# Heavy metal concentrations measured in *P. × generosa* and *S. viminalis* tissues, inoculated (Gi) or non-inoculated (Ctrl) with *G. intraradices*, at the end of the second year of growth.



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Zn	Sv=Pg	Sv>Pg***	Sv>Pg*	Sv>Pg**
Cu	Sv>Pg**	Sv>Pg***	Sv>Pg*	Sv>Pg*
Pb	Sv>Pg***	Sv=Pg	Sv=Pg	Sv=Pg



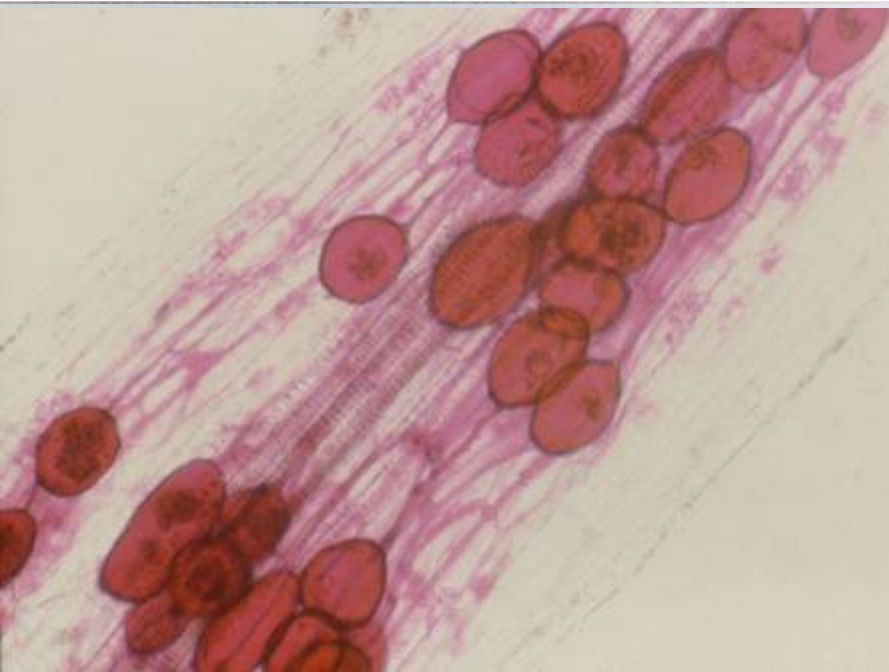
Mean biological concentration factors (BCF) in the leaves and the stems of *P. × generosa* (Pg) and *S. viminalis* (Sv) clones, inoculated (Gi) or non-inoculated (Ctrl) with *G. intraradices*, at the end of the second growing season.

Clones	Tissues	Inoculation	Biological concentration factor <sup>a, b</sup>			
			Cd	Zn	Cu	Pb
<i>P. × generosa</i>	Leaves	Gi	<b>2.56</b>	<b>1.24</b>	0.15	0.014
		Ctrl	<b>3.29</b>	<b>1.35</b>	0.21	0.012
	Stems	Gi	<b>1.87</b>	0.44	0.09	0.030
		Ctrl	<b>2.31</b>	0.46	0.12	0.038
<i>S. viminalis</i>	Leaves	Gi	<b>2.97</b>	<b>1.52</b>	0.22	0.025
		Ctrl	<b>4.41</b>	<b>1.76</b>	0.29	0.037
	Stems	Gi	<b>3.30</b>	<b>1.12</b>	0.19	0.083
		Ctrl	<b>5.50</b>	<b>1.35</b>	0.24	0.081

a Biological concentration factor = tissue concentration / soil concentration (n=20).

b Values in bold are active bioaccumulation (>1).







**Percentage of root length bearing AM fungi structures of the *S. viminalis* and *P. × generosa* clones, inoculated (Gi) or non-inoculated (Ctrl) with *G. intraradices* at planting, in the surface (0-20 cm) and deep (20-40 cm) soil layers, on the second year of the field trial.**

Clones	Inoculation	Mycorrhizal root colonization (%) <sup>a, b</sup>	
		Surface (0-20 cm)	Deep (20-40 cm)
<i>P. × generosa</i>	Gi	48.9 a	40.0 a
	Ctrl	44.6 a	36.6 a
<i>S. viminalis</i>	Gi	5.8 b	2.8 b
	Ctrl	4.8 b	2.6 b



**Results of paired t-tests comparing *S. viminalis* and *P. x generosa* aboveground plant heavy metal concentration in leaves and stems of inoculated (Gi) and non-inoculated (Ctrl) plants between the first (1) and second (2) year of growth in the field.**

Clones	Tissues	Inoculation	Metal concentration			
			Cd	Zn	Cu	Pb
<i>P. x generosa</i>	Leaves	Gi	1=2	1=2	1=2	1=2
		Ctrl	1=2	1=2	1=2	1=2
	Stems	Gi	1=2	1=2	1=2	1=2
		Ctrl	1=2	1=2	1=2	1=2
<i>S. viminalis</i>	Leaves	Gi	1=2	1=2	1=2	1<2**
		Ctrl	1=2	1=2	1=2	1<2*
	Stems	Gi	1<2**	1<2**	1<2**	1=2
		Ctrl	1<2*	1<2**	1<2*	1=2











# Conclusions

- **Willows and poplars constitute wonderful tools to restore brownfields or polluted sites in urban areas;**
- **Their rapid establishment and growth allow them to quickly create a green cover with positive environmental, economic and social impact...**



# Conclusions cont'd...

- **Willows in particular showed interesting ability to tolerate and absorb large quantities of trace metals (notably Zn and Cd);**
- **Inoculation with arbuscular mycorrhizal is possible and constitutes an interesting approach to increase the establishment, but their impact on absorption capacity has not been demonstrated in the studies conducted.**



# Acknowledgements

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- **Fonds des priorités gouvernementales en sciences et en technologies du ministère de l'Environnement du Québec**
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