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Herbicides and nonylphenol in sewage sludge:

occurrence and ecotoxicological impacts

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Phytopharmacie
Versailles
Xénobiotiques
Toulouse



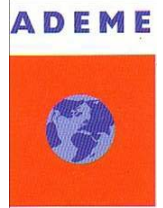
CECOTOX
Lausanne



Santé Publique et
Environnement
Chatenay-Malabry



Biochimie
Paris



Gestion Biologique
des Sols
Angers

Rationale

Scientific

- **Selection of sludge and contaminants**
 - type of process and sludge treatment in WWTP
 - persistent organic pollutants ?
 - analytical methods
 - variation of concentration in sludge with time and WWTP
- **Transfer of contaminants**
 - amounts and forms
 - waters, plants, webfoods
- **Impact of contaminated sludge**
 - soil properties and microbial activity
 - écotoxicology
 - relationships environment / health, EDCs

Socio-economic

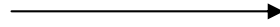
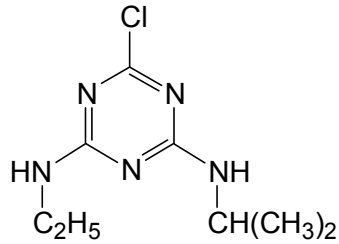
- **Management and sustainability of sludge spreading onto agricultural soils**

Objectives of our research program

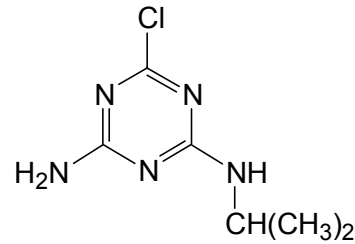
- ***Determine the occurrence of selected pesticides in sludge samples***
 - development of analytical methods
 - annual monitoring in various sludge samples
- ***Identify possible ecotoxicological effects of the pesticides when applied onto the soils in contaminated sludge***
 - fate of the pesticides in the soil
 - ecotoxicity on selected organisms

The chemicals

➤ Atrazine

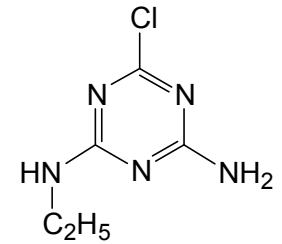


DEA

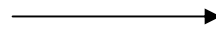
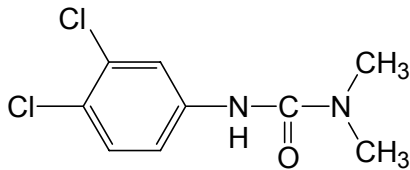


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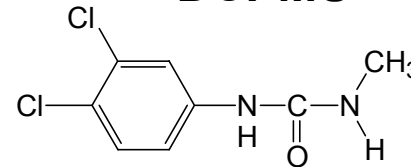
DIA



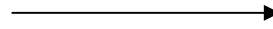
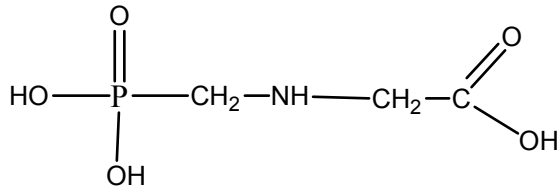
➤ Diuron



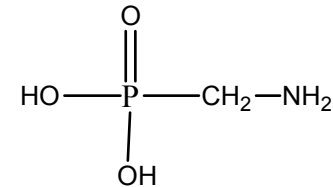
DCPMU



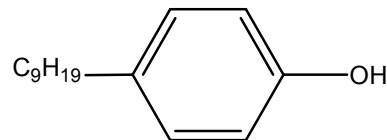
➤ Glyphosate



AMPA



➤ Nonylphenol



1 – Sludge samples contain pesticides

≈ 80 samples from 4 WWTPs during 1 year near Versailles (France)

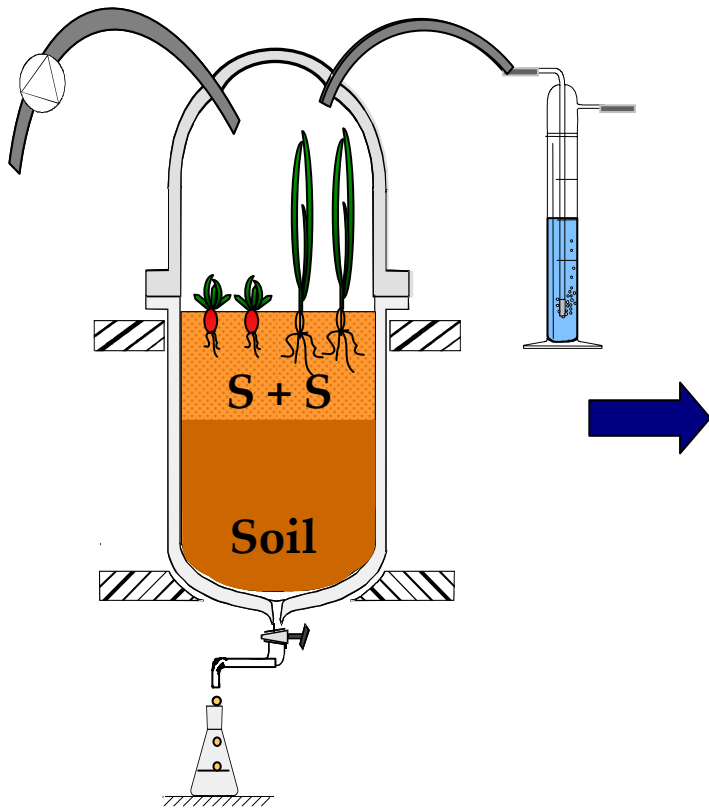
Compound	Dried	Composted	Limed 1	Limed 2
Atrazine + met	/	/	/	/
Diuron ●	11.2	26.5	20.0	46.6
DCPMU	/	/	/	/
Glyphosate ●	1.1	1.0	1.4	0.4
AMPA ●	20.3	12.4	11.5	2.8
Nonylphenol ●	61.7	132.8	90.9	62.5

● ppb ($\mu\text{g}/\text{kg}$ dry sludge) / ● ppm (mg/kg)

French regulation: PCB 0.8 and BaP 2 ppm / EC regulation: nonylphenol 50 ppm

► *It is necessary to study their fate in the soil*

Light 16 h, 21°C / darkness 8 h, 18°C

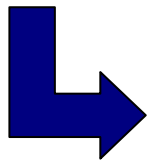


Transformation

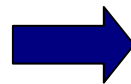
- Mixture of labelled Gly/Diu/NP at 6/17/7 ppm
 - Soil
 - Soil + spiked sludge at 30 and 6 T/ha
- mineralised / extractable / stabilised
metabolic maps

Mobility

- Transfer soil / higher plant

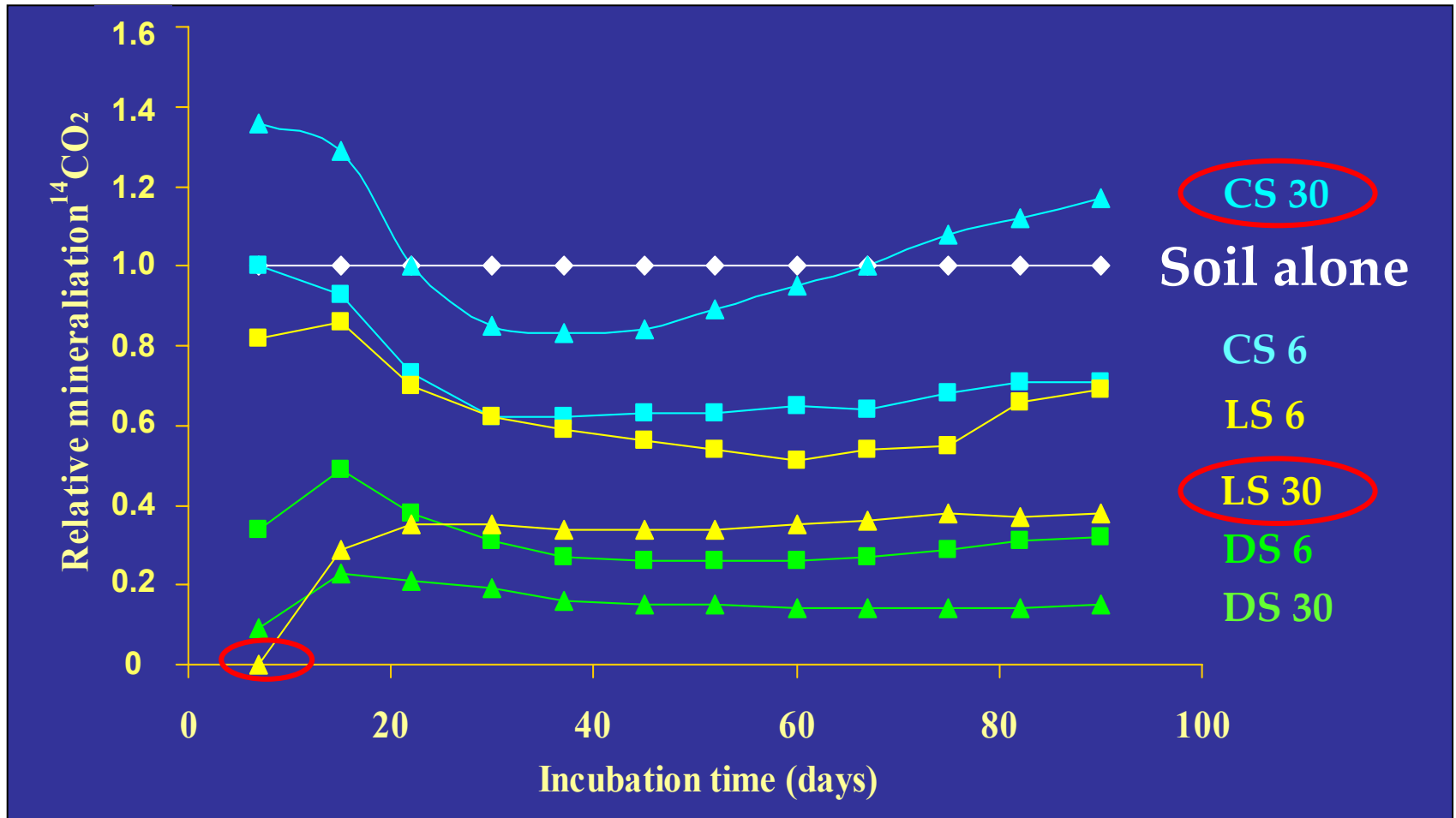


Leachates



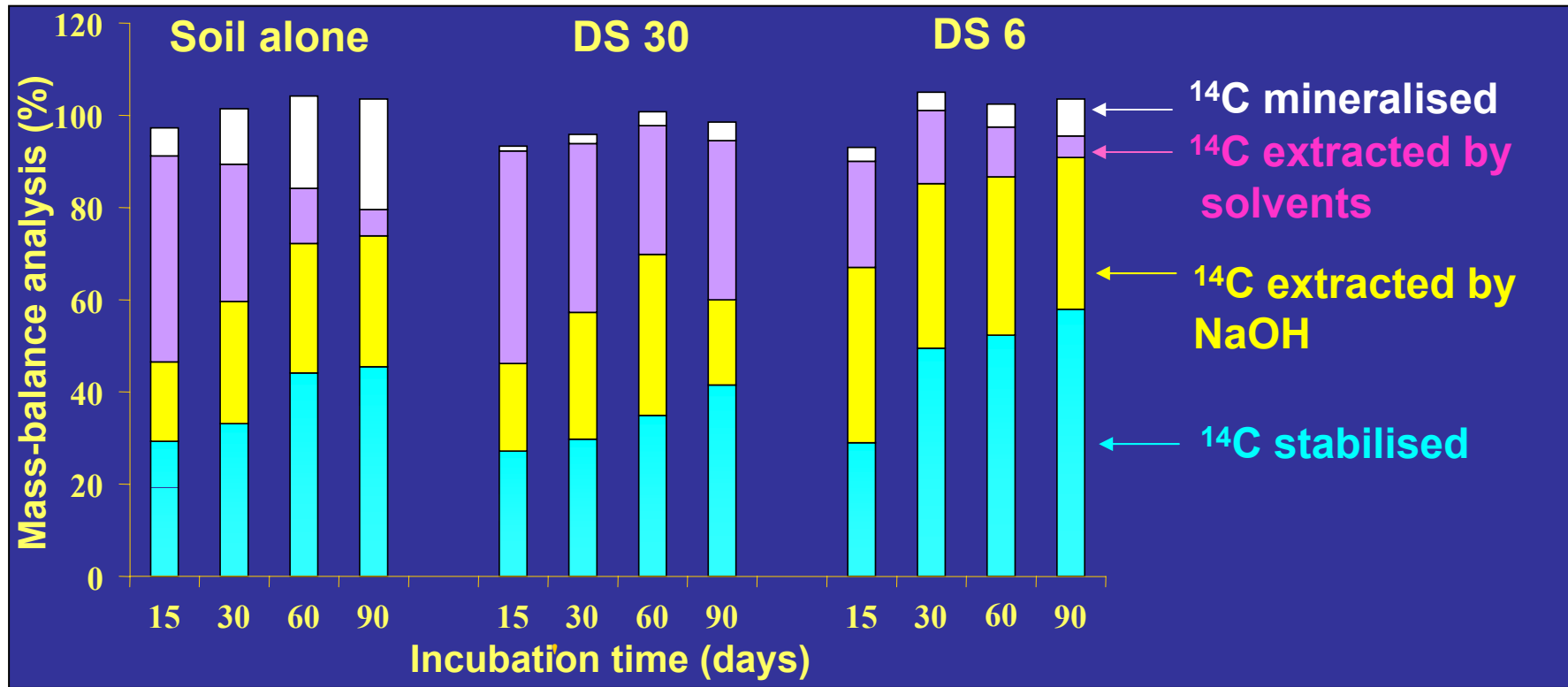
Water contamination

2 – The mineralisation of the pesticides is decreased



- Composted sludge has contrasted effects at high dose
- Limed sludge induces a 7-day lag phase at high dose

3 – The persistence of the pesticides is increased



With time

Mineralisation increased
Extractability decreased
Bound residue increased

With sludge at 30 T/ha

- **Mineralisation decreased**
- **Extractability maintained**

With sludge at 6 T/ha

- **Intermediate situation**

4 – Leachates contain traces of pesticides

Less than 1 % of initial radioactivity with a rainfall of 20 mm

Conditions	D + 45	D + 60	D + 90
	µg/L	µg/L	µg/L
Soil alone	2.1	1.6	1.2
+ Dried sludge (30 T/ha)	2.5	0.9	1.2
+ Dried sludge (6 T/ha)	0.5	0.3	0.2
Soil alone	14.5	16.2	11.0
+ Composted sludge (30 T/ha)	14.9	12.5	15.2
+ Composted sludge (6 T/ha)	4.8	5.2	4.7
Soil alone	14.5	16.2	11.0
+ Limed sludge (30 T/ha)	29.6	25.2	25.4
+ Limed sludge (6 T/ha)	3.4	3.8	3.7

- Nonylphenol is the main compound detected using HPLC

5 – Sludge modify the transfer of pesticides from the soil to higher plants

Less than 0.05 % of the radioactivity during the period of growth

Conditions	Radish (15 days) µg/g	Wheat (45 days) µg/g
Soil alone	/	0.9
+ Dried sludge (30 T/ha)	/	0.07
+ Dried sludge (6 T/ha)	/	/
Soil alone	6.0	0.7
+ Composted sludge (30 T/ha)	1.1	1.2
+ Composted sludge (6 T/ha)	1.8	1.6
Soil alone	6.0	0.7
+ Limed sludge (30 T/ha)	4.3	0.1
+ Limed sludge (6 T/ha)	/	1.1

It is necessary to study their ecotoxicological impact



Assays including 3 ecological levels

- **Degraders**

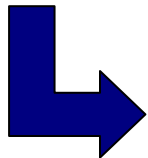
- microbial functional diversity
« litter bag »
 - structural diversity
PLFA

- **Producers**

- germination (% , delay) of plants
biomass

- **Consumers**

- mortality, reproduction of springtails

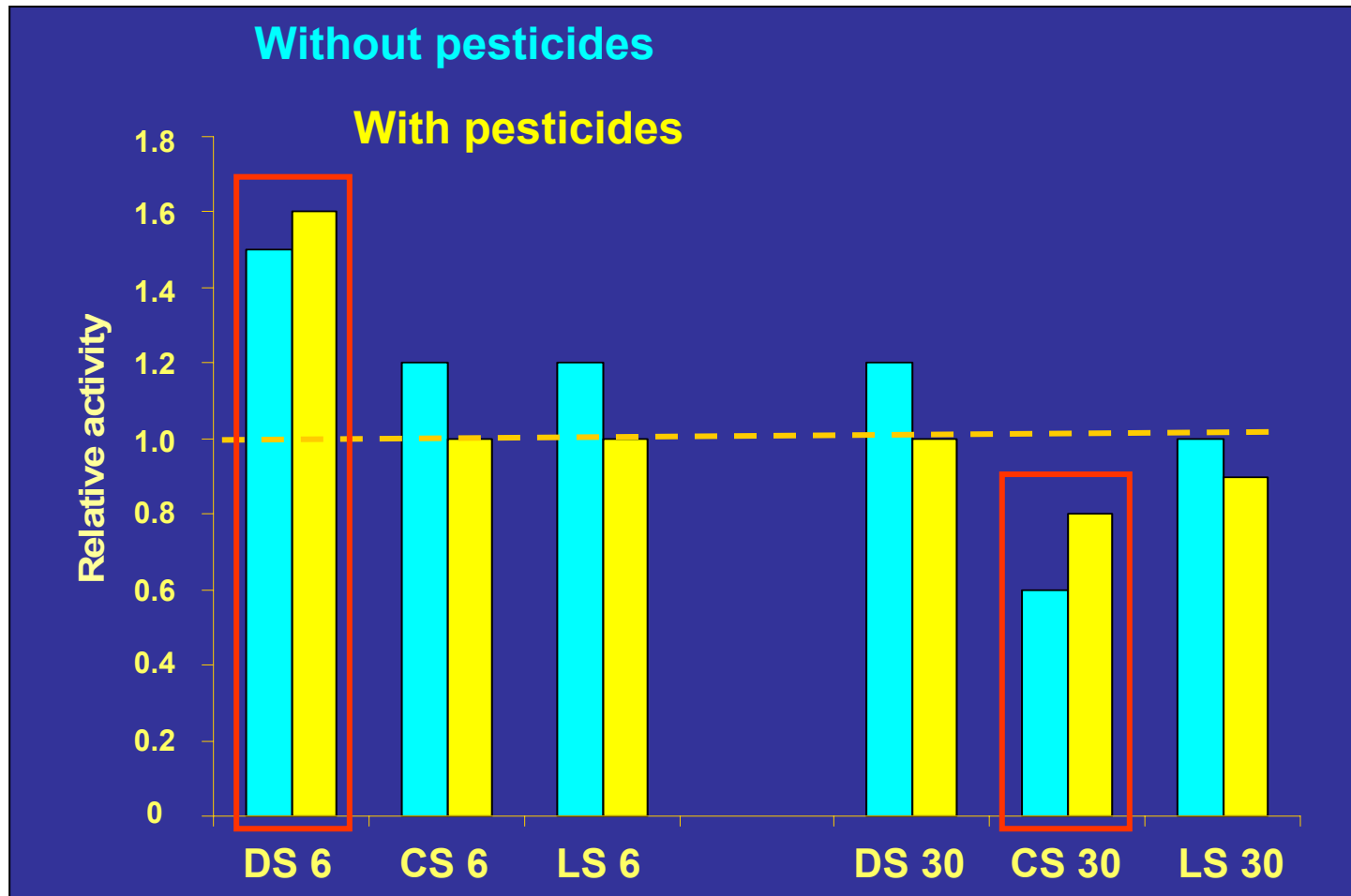


Estrogenicity assessment
human cell lines MELN



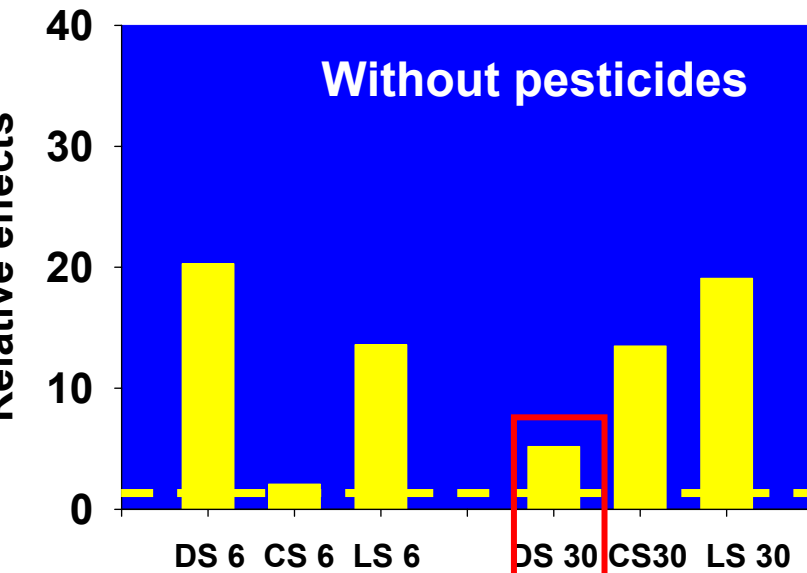
Risk for humans

6 – Pesticides have no clear impact on the litter-bag

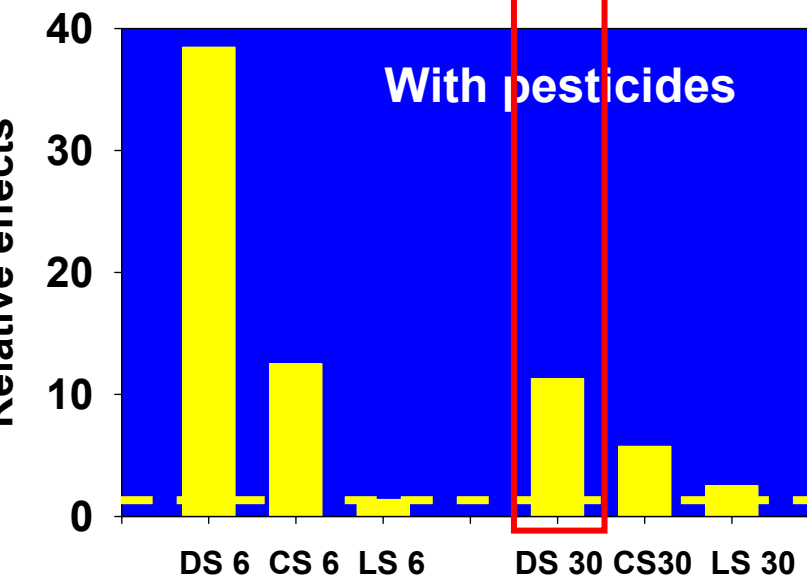


- Dried sludge increases the soil biological activity at low dose
- Composted sludge decreases the soil activity at high dose

7 – The fungi/bacteria ratio is increased

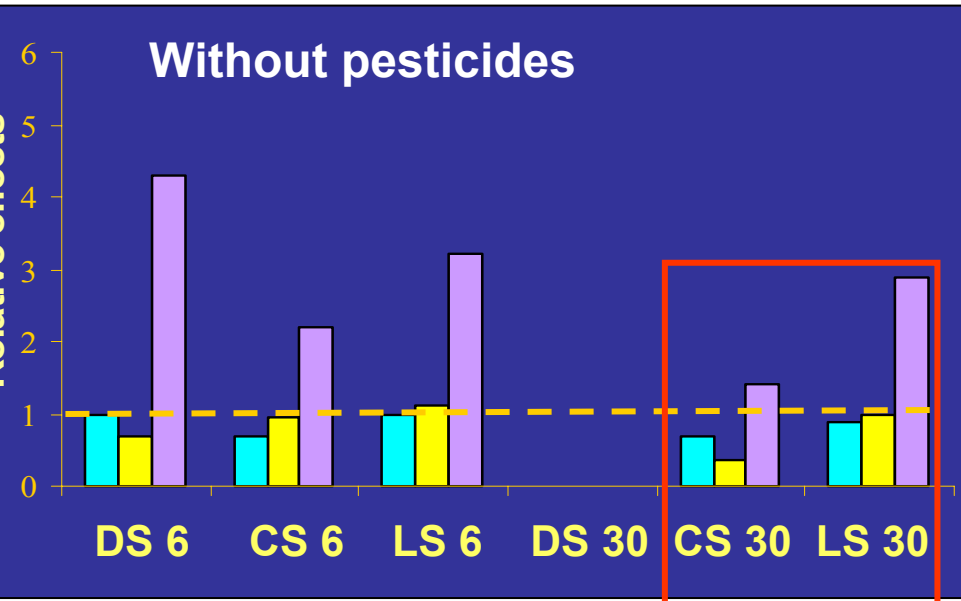


- Sludge intake generally increases the fungi/bacteria ratio
- The highest effects are noticed with the dried sludge at low dose
- There is no clear effect of pesticides

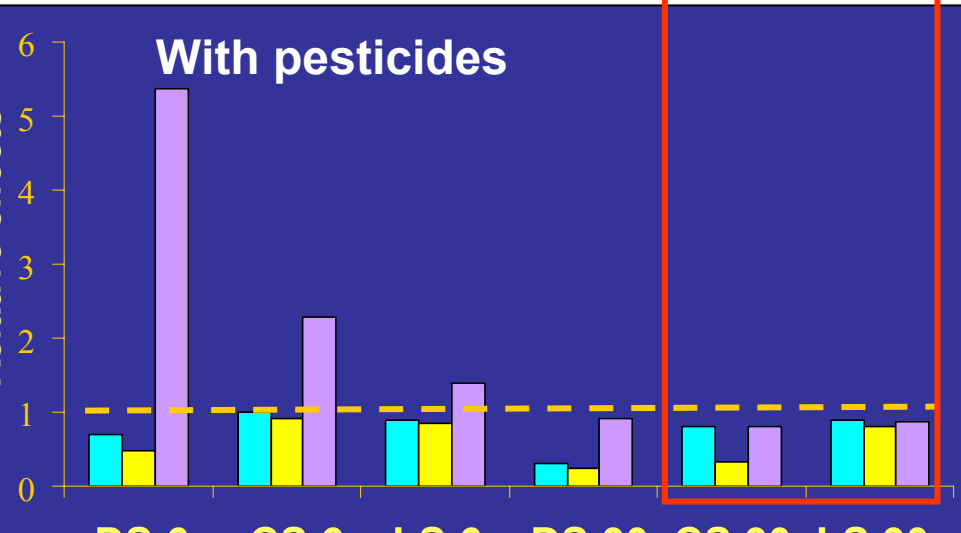


- Dried sludge at high dose induces the development of fungi at the soil surface

8 – Sludge are phytotoxic on wheat at high doses



% of germination, delay, biomass production



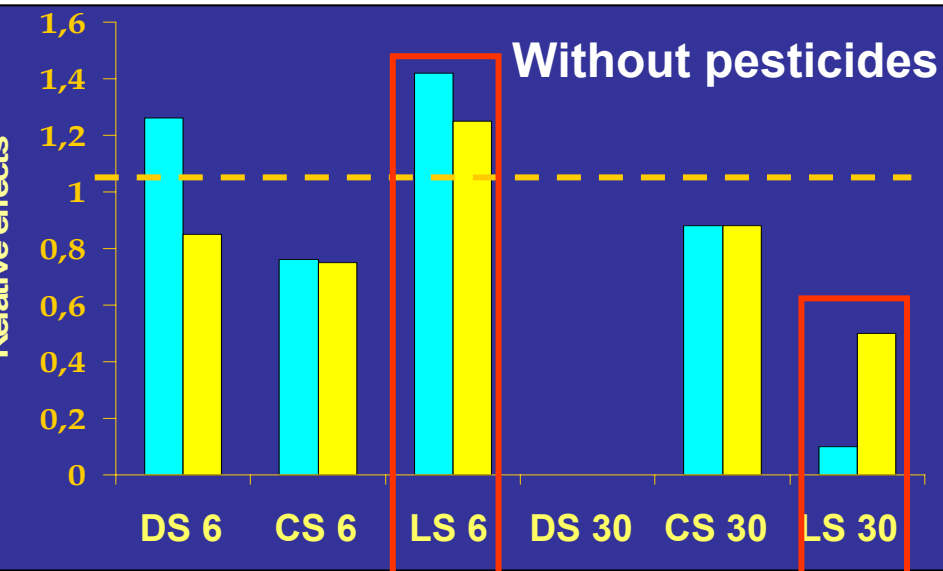
- Sludge are fertilizers at low dose
- Dried sludge inhibits plant development at high dose (fungal proliferation)

- Spiked sludges are phytotoxic at high dose
- Phytotoxicity could be due to pesticides

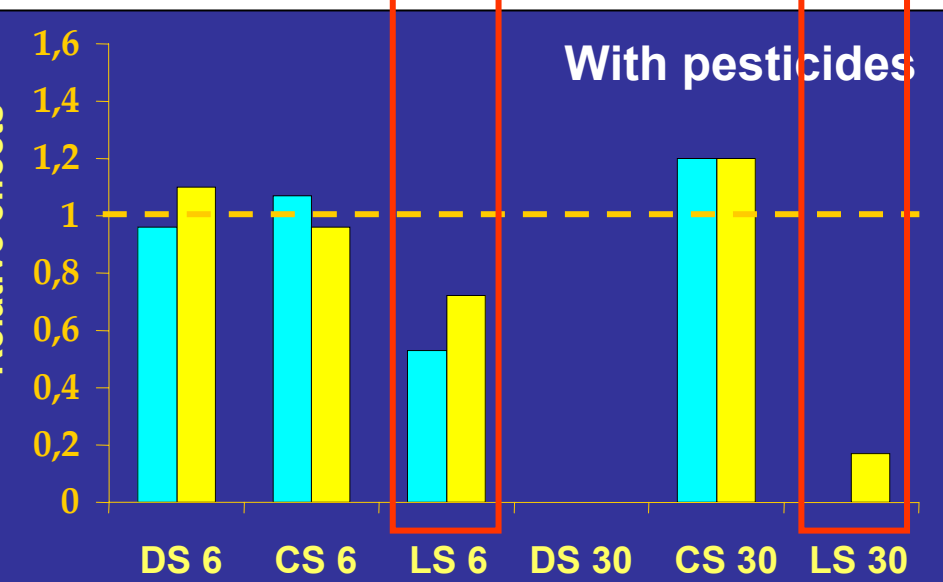
9 – Sludge are toxic for springtails



F. candida



Reproduction survival

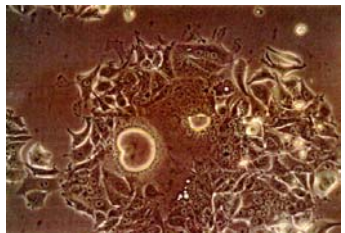


- No effect at low dose
- Dried sludge inhibits springtails life at high dose (fungal proliferation)
- Limed sludge inhibits springtails survival at high dose (pH?)

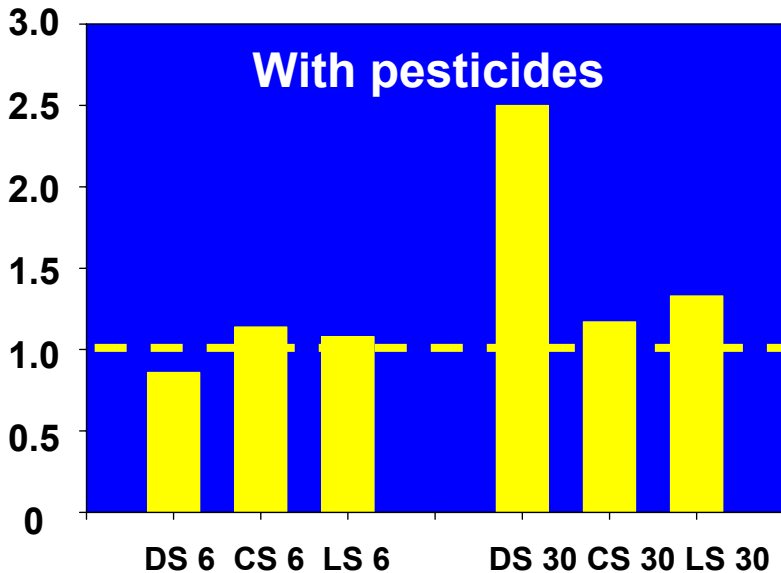
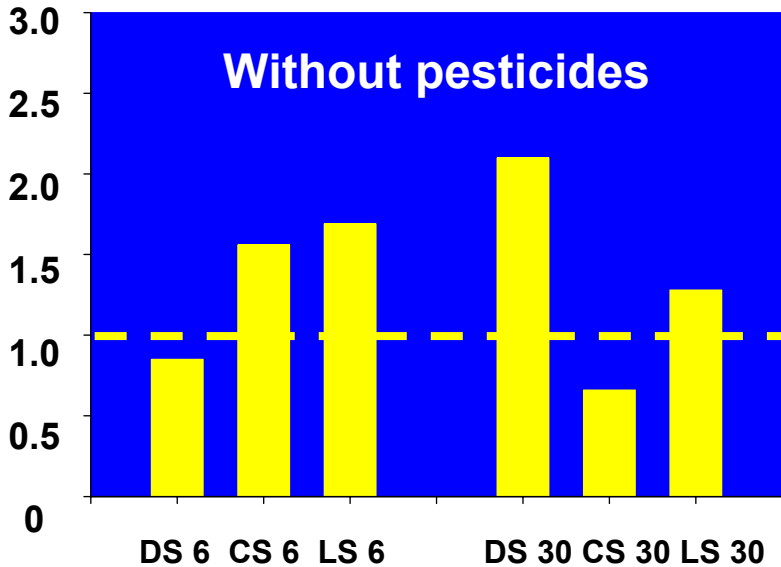
- Limed sludge strongly inhibits springtail survival at high dose

- Toxicity could be due to pesticides

10 – Leachates induce no clear estrogenic effects



MELN cells



- Dried sludge only induced significant estrogenic effects on cells at high dose
- That effect seems strengthened by pesticides (nonylphenol ?)

Conclusions and perspectives

- Urban sewage sludge contain pesticides at relatively high concentrations
- The persistence of pesticides entering the soil through sludge is increased
- Eco-toxic effects have been evidenced at the high dose of sludge (worst case), and not in an agronomic reality
- The effects of sludge are in general higher than these of added pesticides
- The process in WWTPs must be improved to degrade organic pollutants

Acknowledgements

- **The colleagues of the 4 research institutes**
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A D E M E

