



### Behavior canola (Brassica Napus) following a sewage sludge treatment

Najla Lassoued, Essaïd Bilal, Saloua Rejeb, Issam Guénole-Bilal, Naceur Khelil, Mohamed Nejib Rejeb, Frédéric Gallice

### ► To cite this version:

Najla Lassoued, Essaïd Bilal, Saloua Rejeb, Issam Guénole-Bilal, Naceur Khelil, et al.. Behavior canola (Brassica Napus) following a sewage sludge treatment. Carpathian Journal of Earth and Environmental Sciences, North University Center of Baia Mare, Romania, 2013, 8 (3), pp. 155-165. <hl>

### HAL Id: hal-00904278 https://hal.archives-ouvertes.fr/hal-00904278

Submitted on 14 Nov 2013  $\,$ 

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers. L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

### BEHAVIOR CANOLA (BRASSICA NAPUS) FOLLOWING A SEWAGE SLUDGE TREATMENT

# Najla LASSOUED<sup>1,2</sup>, Essaid BILAL<sup>3</sup>, Saloua REJEB<sup>2</sup>, Issam GUENOLE-BILAL<sup>4</sup>, Mohamed Naceur KHELIL<sup>2</sup>, Mohamed Nejib REJEB<sup>2</sup>, & Frédéric GALLICE<sup>3</sup>

 National Agronomic Institute of Tunisia, 1082 Mahragène Tunis, Tunisia; lassoued\_najla@yahoo.fr 2 National Institute for Rural Engineering' research, Water and Forestry, BP10, 2080 Ariana, Tunis
3 Ecole Nationale Supérieure des Mines de Saint Etienne, GSE, CNRS UMR 5600, F42023 Saint Etienne France; bilal@emse.fr

4 Physiology Department, Lausanne-CHUV University, Lausanne, Switzerland; bilal.issam.1698 @ gmail.com

Abstract: In this study, two types of sludge were being used, while the first was with urban dominance, the second was with industrial dominance. The effects of sewage sludge had been studied in a Brassica Napus field. The mineralogical, chemical and microscopic study of the sludge showed that industrial sludge had very high levels of Cr, Pb and Cd. These metals existed mainly under the form of daubreelita Cr<sub>2</sub>FeS<sub>4</sub>, brezininaite Cr<sub>3</sub>S<sub>4</sub>, wattersite Hg<sub>5</sub>CrO<sub>6</sub>, crocoite PbC<sub>2</sub>O<sub>4</sub>, pheonicochroite Pb<sub>2</sub>O(CrO<sub>4</sub>) and Pboxalate PbC<sub>2</sub>O<sub>4</sub>. The results showed that sludge significantly improves the growth of the underground part of the plant (root) and the upper part (stem, leaves etc...). This improvement is more important for urban sludge. However, this beneficial effect was accompanied by a change in the composition of the plant some trace element metals. An abnormal accumulation of Cr was found in the roots, stem, leaves, and siliques when the industrial sludge was brought which reflected the richness of the latest. The doseeffect sewage sludge was very clear at the levels of Pb in the roots especially for industrial sludge which exceeded the threshold values of toxicity starting from the dose of 25t/ha of industrial sludge. Cd levels only increased with the addition of 100t/ha of industrial sludge. For Ni, Cu, Co and Zn, especially at roots level, the increase depends on mud's dose and especially on its type. On the contrary, levels of iron, and to a less extent manganese levels, had been reduced due to sludge despite their richness with these elements. That was probably due to antagonism with one or more particular elements especially Zn.

Key words: heavy metals, accumulation, Brassica Napus, urban sludge, industrial sludge.

#### **1. INTRODUCTION**

In Tunisia, the amount of sludge produced by wastewater treatment stations is constantly increasing, while ways for the disposal and recovery are not progressing. The agricultural use of sludge is highly demanded and not only spreading it on soil seems to be an alternative solution that could be acceptable, but also it looks like the least expensive recycling solution. This practice allows on one hand to recycle sludge and enjoy its fertilizing properties by providing nutrients such as nitrogen, phosphorus, magnesium, etc. and on the other hand it permits the closure of the cycle by returning organic material to the ground.

Among the sludge, there are urban sludge and industrial sludge. The first is produced in urban waste water treatment stations, in other words, most of it is wastewater from domestic sources. The second type is obtained from the treatment of industrial wastewater or partly from industrial wastewater. When purifying wastewater, heavy metals accumulate in the sludge. Thus, the sludge contains significant amounts of these elements and particularly when it is a case of industrial sludge (Juste et al., 1995; Rejeb et al., 1999, Nicholson et al., 2003). These amounts of sludge are sources of contaminants containing many organic components, well vis-à-vis metals. Among the main pollutants generated by industrial activities, heavy metals (Cu, Pb, Cr) actually raise issues of particular concern. These non-biodegradable components have high ecological toxicity and may be involved in many diseases (infections of the central nervous system, liver, kidneys, as well as cancers and embryonic malformations (Abrahams, 2002; Adriano, 2001).

Plants are the major raison of the presence of metals in the food chain. Their concentration into trace elements, in contexts diffuse contamination is highly variable. For a given level of contamination of an environment, the accumulation depends not only on the component, the plant species' family and variety, but also on the body and soil factors such as pH, temperature, (Kuboi et al., 1986; Coullery, 1997; Rejeb et al., 2011).

In this context, we undertook this study to understand the physiology of *Brassica napus* L., belonging to the family *Brassicaceae*. This large biomass plant and a deep root system is recognized as growing in metal accumulator, being a good candidate for induced phytoextraction (Veerle Grispen et al., 2006; Sellami et al., 2012; Ben Ghnaya et al., 2009).

#### 2. MATERIAL AND METHOD

#### **2.1. Experimental Protocol**

The test is to show the effect of two types of sludge, urban and industrial, on the behavior of Canola (*Brassica napus*). The sludge doses (5, 25, 50 and 100 t/ha) in soil had been compared to a soil without any input.

The experimental protocol had been installed in the field at the Agricultural Experiment Station of Oued Souhil - Nabeul, located about 60 km from Tunis and belonging to the National Institute for Rural Engineering Water and forest. The site is characterized by a semi-arid superior bioclimatic zone. During this test the changes in temperature and rainfall were followed. For the region of Nabeul, the average annual temperature under cover is 19.2°C. However, the maximum average temperature of the hottest month for 10 years was 26.5°C (August) and that of the coldest month is 10.5°C (February). The annual rainfall varies from 390 to 630 mm/year. Sand is the most representative size fraction of the soil of the test plot. Depending on the texture triangle, the studied soil is a sandy loam soil. The analytical results show that our soil is characterized by an alkaline pH (Table 1), electrical conductivity varies from 0.42 to 0.89 mS/cm indicating a low salinity due to the sandy nature of the soil. Regarding the total limestone, our soil has a low rate below 5%. The organic matter content is low in the range from 0.39% to 1.98%.

The experimental plot with an area of 750 m<sup>2</sup> ( $25m \times 30m$ ) was divided into 36 basic plots of 10 m<sup>2</sup> ( $5m \times 2m$ ) separated by a neutral zone of 1.5m. The test is conducted in four randomized blocks with 9 treatments (T: soil without sludge contribution and

soil with 8 treatments of 5, 25, 50 and 100 t / ha urban (BU) and industrial (BI) sludge respectively).

Urban sludge was taken from the wastewater treatment plant of Korba station with a low load activated sludge treatment system followed by maturation. The sludge from the station had been under aerobic stabilization followed by drying beds. The dry sludge was removed from the drying bed. The other type was the industrial sludge taken from Bou Argoub sewage treatment plant hosting two industrial zones, companies in the Refrigeration Company and brewery Tunis (SFBT) specialized in the food industry, and Assad specialized in the electrical industry. The sludge from the station had been under an aerobic stabilization followed by drying beds. This sludge was containing heavy metals especially Pb and Cr.

Table1. Chemical composition and characteristic of soil was used for quality control. All these values are the averages of four replicates.

Soil control						
pН		$8.36 \pm 0.51$				
Humidity	%	$7.4 \pm 1.62$				
CE	ms/cm	$0.61\pm0.18$				
С		$0.34\pm0.05$				
MO	%	$0.64\pm0.09$				
Ν		$0.045\pm0.004$				
C/N		$7.55 \pm 0.88$				
LOI		$5.355 \pm 1.02$				
SiO <sub>2</sub>		$87.12 \pm 11.2$				
TiO <sub>2</sub>		$0.21\pm0.02$				
$Al_2O_3$		$1.91\pm0.25$				
$Fe_2O_3$		$0.95\pm0.11$				
MnO	%	$0.02 \pm 0.003$				
MgO		$0.26\pm0.09$				
CaO		$1.55 \pm 0.2$				
Na <sub>2</sub> O		$0.02\pm0.001$				
K <sub>2</sub> O		$0.62 \pm 0.2$				
$P_2O_5$		$0.2 \pm 0.06$				
Cd		trace				
Co	ppm	$0.6101\pm0.19$				
Cr		$2\overline{1.381 \pm 4.3}$				
Cu		$2\overline{5.805 \pm 5.9}$				
Ni		$3.4099\pm0.49$				
Pb		$16.589 \pm 2.46$				
Zn		$70.502 \pm 11.06$				

The slurry was mixed thoroughly and spread manually on the ground 30 days before planting. Rape seed were sown at a depth of 5 cm with a spacing of 40 cm and cm interval on the line with about 50 seeds /  $m^2$ . During the test, the water needs of the crop were determined using Cropwat 4.3 software (Savva & Karen, 2002) based on the Penman - Monteith changed.

## 2.2. Chemical analysis of wastewater treatment sludge and soil

For sewage sludge and soil, the pH was measured in 1 M KCl after 24 h in water / soil ratio of 5. The organic carbon was determined by Kalra & Maynard method's (1991). The total nitrogen (NT) was determined by Kjeldahl method. For trace elements, samples of soil and sludge were digested with a mixture of HCl/HNO<sup>3-</sup> (McGrath & Cunliffe, 1985) and total concentrations were determined by ICP-AES (Inductive Coupled Plasma Atomic Emission Activa-Horiba Spectrometry Jobin Yvon Spectrometer). The major elements were determined by X-ray fluorescence with a preparation under the form of pearl borate. The device used was a fluorescence spectrometer dispersive X length Siemens Bruker (SRS3400). The all analysis had realized in the Geosciences and environment Department of Ecole Nationale Supérieure des Mines de Saint Etienne.

#### 2.3. Mineralogical analysis

Mineralogical analysis was performed by Xray diffractometry. The equipment used was a Siemens D5000 diffractometer comprising a vertical goniometer / with a rotating sample holder, a smuggler of 40 positions, rear graphite monochromator and a scintillation counter. The operating system was computerized allowing the programming of data acquisition operations. The crystalline fraction of the samples was determined by XRD from a powder pattern. The proportions were estimated by peak areas.

The urban and industrial are studied by Scanning Electron Microscopy (SEM) JSM-6400 associated with a microanalysis device of energy dispersive spectrometry X-ray.

#### 2.4. Control parameters at plant level

Canola harvesting was carried out after the formation of siliques at the end of May 2011. The plants were separated into young pods, leaves, stems and roots. The roots were soaked for 5 min in cold 0.01M HCl solution to remove any heavy metals adsorbed on the surface of the root, then rinsed three times with distilled water (Aldrich et al., 2003) and dried with filter paper. The fresh weight was measured immediately, and dry weight after 48 h of drying in an oven at 60 °C.

#### • Foliar area measurement

This measurement was performed on all the leaves of the plant at a sufficiently advanced stage of development but still before the loss of basal leaves. Leaf area was measured using a planimeter Area Mesurement System (Conveyor Blet unit 230V/50Hz) coupled to a computer with a WINDAS system (Colour Image Analysis System).

#### • Trace metal dosage in the plant

Digestion of plant samples was performed using hot nitric concentrated acid, according to Zarcinas et al., (1987), Petrescu & Bilal (2003, 2006. 2007), Secu et al. (2008) et Lăcătușu et al. (2009). Samples of dried plants were ground into powder using a porcelain mortar and pestle to get a fine powder. An amount of 500 mg of this plant powder was digested with a mixture of 4 ml HNO<sup>3-</sup> to a temperature of 70 °C for 1h 50mn. After a 12 hour rest, 3ml of H<sub>2</sub>O<sub>2</sub> were added. Then, it was heated at 70°C to collect the remaining residue. To the latter 2 ml and 4 ml of HNO<sub>3</sub>-HCl were added to form aqua regia. Then, the contents of heavy metals (Pb, Mn, Zn) were determined by emission spectroscopy plasma torch (ICP-AES) HORIBA Jobin Yvon) in the Department Geosciences and Environment of Ecole Nationale Supérieure des Mines de Saint Etienne.

#### 2.6. Statistical Analysis

To confirm the data variability and results validity, all data were subjected to variance analysis. The comparisons of averages at the threshold of 5% significance were made by the Newman-Keuls test using the Statistica 7 software.

#### **3. RESULTS**

# **3.1.** Mineralogical, chemical and physical study of urban and industrial sewage sludge

The different studied characteristics (pH, humidity, carbon and heavy metal) of sludge are shown in Table 2.

Table 2 shows that the pH of the sludge is low (<7), which promotes the release of metals. An acid pH causes the metal salts in solution, the dissolution of the retention phases, desorption of cations and adsorption of anions.

However, this sludge contains organic matter in significant proportions (up to 66%) which promotes their use in agriculture. The C/N ratio defines the mineralization potential. The less it is, the faster is the mineralization. C/ N of sludge used varied from 7.4 to 7.5.

The main elements present in the major sludge are: silica, alumina and at second level, the iron oxide and potassium oxide. The presence of high levels of CaO is due to the use of lime in the treatment of sludge for a better cleaning and improved odor control (Barbe et al., 2002).

Regarding metal traces, chemical analysis of urban sludge of Korba (Table 2) showed that Fe and Zn were the most represented elements. The average levels found were organized according to the following sequence: Zn > Cu > Cr > Pb > Ni > Co >Cd. As for industrial sludge of Bou Argoub, it had very high levels of Cr, Pb and Cd. The found sequence was Cr >>>Pb>>Zn> Cd>Cu>Ni>Co.

Table 2: Chemical composition and characteristic of the urban (BU) and industrial (BI) sewage sludge. All these values are the averages of four replicates.

		BU	BI		
pH		$6.70\pm0.25$	$6.3\pm0.55$		
Humidity	%	$42.7\pm3.25$	$39.5\pm3.5$		
CE	ms/cm	$8.37 \pm 1.6$	$9.7\pm1.03$		
С		$38{,}96\pm7.2$	$31.88 \pm 5.2$		
MO	%	$66.62 \pm 6.5$	$57.93 \pm 4.2$		
Ν		$5.2 \pm 1.3$	$4.3\pm0.9$		
C/N		$7.49 \pm 1.5$	$7.41\pm0.75$		
LOI		$52.305\pm8.6$	$52.145\pm8.2$		
SiO <sub>2</sub>		$20.71\pm3.5$	$28.56 \pm 2.5$		
TiO <sub>2</sub>		$0.27\pm0.07$	$0.13\pm0.02$		
$Al_2O_3$		$4.41 \pm 1.2$	$1.91 \pm 0.4$		
Fe <sub>2</sub> O <sub>3</sub>		$1.88\pm0.5$	$1.02\pm0.2$		
MnO	%	$0.02\pm0.002$	$0.03\pm0.001$		
MgO		$1.22\pm0.2$	$0.8\pm0.09$		
CaO		$13.41\pm2.57$	$8.51 \pm 1.87$		
Na <sub>2</sub> O		$0.33\pm0.09$	$0.44\pm0.07$		
K <sub>2</sub> O		$0.84 \pm 0.1$	$0.36\pm0.05$		
$P_2O_5$		$3.24\pm0.5$	$2.18\pm0.2$		
Cd		-	$163.04\pm11.2$		
Co		$3.6099 \pm 1.1$	$20.614\pm3.51$		
Cr		$73.119\pm9.2$	$11387\pm450.9$		
Cu	ppm	$188.78\pm24.8$	$142.42 \pm 25.3$		
Ni		$19.799 \pm 3.5$	$61.842 \pm 14.6$		
Pb		$54.886 \pm 9.5$	$2854.1 \pm 102.3$		
Zn		463.21±34.9	$821.75 \pm 60.4$		

The observation with a scanning electron microscope of the sludge (Fig. 1) shows an amorphous phase in the form of flakes whose geometry is poorly defined and trapping small crystals of quartz and muscovite.

In this picture, the voids are shown in black, light gray ranges correspond to quartz grains and heterogeneous beaches in dark gray correspond to the clay phase. We determined the mineral composition of the various selected points (Fig. 1). This analysis, revealed the presence of Pb and Cr in A, B, C and D points of industrial sludge (SEM image B Fig. 1) unlike urban sludge where these elements were not present. These results confirm the chemical analysis. These observations correlate with the main mineral phases identified by X-ray diffraction. The spectral analysis can show that the sludge is composed mainly of quartz, gypsum and calcite.



Figure1. SEM image of sewage sludge (A: Urban Sludge and B: Industrial sludge)

The Cr and Pb in the industrial sludge exist primarily in the form of daubreelita  $Cr_2FeS_4$ , brezininaite  $Cr_3S_4$ , wattersite  $Hg_5CrO_6$ , crocoite  $PbC_2O_4$ , pheonicochroite  $Pb_2O(CrO_4)$  and Pb-oxalate  $PbC_2O_4$ Whereas in urban sludge, we noted the lack of Cr and the presence of Pb is in the form of macphersonite  $Pb_4(CO_3)_2(SO_4)$  and lanarkite  $Pb_2O(SO_4)$ .

# **3.2.** Effect of sewage sludge on the production of biomass of Colza

At the end of the run, we measured the leaf surface. We could notice that for the same dose

applied, no type or dose effect of mud sludge on the leaf surface of younger leaves (No. 12) was observed. However, the older leaves (No. 1) showed a significant increase in the contribution of dice area 5t/ha sludge (Fig. 2), this effect is clearer in urban sludge as industrial sludge. This may be due to the richness of urban sludge with nitrogen and the lower trace element load.



Figure 2. The total Canola leaf area linked of different inputs sludge and control soil without treatment.

The results showed that the bringing of urban sludge 50t/ha significantly increased biomass production of root and stem of Colza compared into the control soil (Fig. 3). This effect also occurs at the level of leaves starting from the addition of 5t/ha sludge. An increase in the production of pods was recorded from 25t/ha sludge. For industrial sludge, increased root mass and silique mass was recorded with 100t/ha dose. There is also a significant dose effect of mud on the stem at doses 50 and 100t/ha. For leaves, the effect is visible from 5t/ha. These increases are significantly greater for urban sludge.

At the end of culture, we weighed the weight of seed produced per square meter, for all treatments. The contribution of sludge stimulated seed production (Fig. 4) and lowest production was recorded with treatment without addition of sludge. On the contrary, the contribution of mud had no effect on the WTS weight of a thousand seeds (Fig. 5). The increase in seed production was due to an increase in the number of seed but not the weight of the seed. Otherwise, the contribution of mud significantly improved the nitrogen of the seed (Fig. 4).

This improvement was most noticeable with the contribution of urban sludge and could be related to essential nutrients such as nitrogen and phosphorus supplied by the sludge.

## **3.3.** Accumulation of heavy metals in different parts of the Canola

The analysis of the plant showed that Cd levels are only detectable for treatments with industrial sludge from 25t/ha BI and this in leaves, stems and roots.



Figure 3: Impact of the urban (BU) and industrial (BI) the sewage sludge doses on the evolution of the mass of dry weight in different parts of Canola (Root, Stem, Leaf and silique). Each value represents the average of four individual



treatment

Figure 5. The One Thousand Seed Weight contents in Canola grown in the presence of increasing doses of sewage sludge.



Figure 6. The N contents in Canola grown in the presence of increasing doses of sewage sludge.

At the level of these organs, dose sludge effect is clear. For siliques, no effect was observed (Fig. 7). At the seeds of 100t/ha treatment industrial sludge, some Cd levels exceeded the acceptable threshold according to Mench & Baize (2004).

For both types of sludge, the accumulation of Cr in the various parts of the plant was increasing according to the doses of sludge (Fig. 8). However, this increase was very important for industrial sludge and especially at the roots where concentrations reached 1093 ppm with 100t/ha dose. This was due to the abundance of Cr in the sludge that was on the order of 11 ppm. These values exceeded the natural levels found in plants and confirmed the status of hyper accumulator Canola.



Figure 7. The Cd content in Canola plants grown linked of the presence of increasing doses of urban and industrial sludge.

It is clear from figure 9 for the different treatments; the average levels of Pb in the aerial parts were located below the threshold of toxicity to plants (30 ppm) even industrial sludge. The observed increases with the addition of sludge were small. It was important to note that the level of Pb was at the state of trace and was detectable only for treatments 50 and 100 t/ha of industrial sludge. On the contrary, at the roots level, the dose effect of sludge was very clear especially for industrial sludge which exceeded the threshold values with toxicity starting from the dose of 25t / ha of industrial sludge (74 ppm).

The Zn is accumulated at the root, leaf and seed (Fig. 10). The highest amounts of Zn were obtained for 100t/ha sludge including industrial sludge. For example, leaf analysis revealed a maximum of 254 ppm obtained 100t/ha industrial sludge while with 100t/ha urban sludge, values were recorded in half (139 ppm).

The addition of sludge slightly increased the Ni content of the aerial parts of the plant (leaves, stems, siliques and seeds) for high doses. But remained below extreme values (10 ppm) faced in plants according to Kabata Pendias & Pendias in 1992. However, a good clear dose effect was noted for root parts (Fig. 10). For Ni, the two types of sludge had similar effects and gave equivalent values. The addition sludge increased significantly the Co content in the roots (Fig. 11).





Figure 8. The Cr content in canola plants grown linked of the presence of increasing doses of urban and industrial sludge treatment.



Figure 9. The Pb content in canola plants grown linked of the presence of increasing doses of urban and industrial sludge treatment.

Table 3. The variation of Fe and Mn contents in the leaves, stems, siliques and roots of Colza linked with urban and industrial sludge treatment. The values of the same fabric and the same line between treatments with the same letter are not significantly different at P < 0.05 (Newman-Keuls test).

		Control	5BI	25BI	50BI	100BI	5BU	25BU	50BU	100BU
Root	Fe <sup>2+</sup>	2293 a	2221 a	1948 ab	1527 b	1485 b	2264 a	2242a	2190 a	2208 a
	Mn <sup>2+</sup>	23.25 a	20.5 ab	17.75 ab	16.75 ab	15.75b	23.5 a	21.5 a	20.25 a	19.25 a
Stem	Fe <sup>2+</sup>	886 a	691 b	619 b c	599 b c	432 c	648 a	703 a	695 a	551 a
	Mn <sup>2+</sup>	22.75 a	23 a	21.5a	22 a	20.5 b	19 a	18.5 a	18.5 a	18.75 a
Siliques	Fe <sup>2+</sup>	1400 a	1016 b	971 b	945b	928 b	992ab	658ab	1114 b	707 b
	Mn <sup>2+</sup>	37.75 a	31 b	31.25 b	32.5b	29.5b	33.25 b	32b	30.25 b	31 b
Leave	Fe <sup>2+</sup>	419.3 a	410.5a	375.8 a	358.8 a	284.6b	409.0a	355.3ab	313.8 ab	312.0 b
	Mn <sup>2+</sup>	117.3 a	96 b	89.8 b	89.8 b	70.4 c	85.8 b	85.0 b	81.8 b	70.3 b



Figure 10. The Zn contents in Canola grown in the presence of increasing doses of sewage sludge. T: control soil



Figure 11. The Ni contents in Colza plants grown linked of the presence of increasing doses of urban (BU) and industrial (BI) sludge. Témoin: control soil.

The Co contents in the leaves also increased with the addition of sludge but the dose effect was not present (Fig. 12). The Cu is increased at the levels of roots, leaves and seeds with input from urban and industrial sludge (Fig. 13). The Fe content decreased significantly in the roots, stems, siliques and leaves with the addition of industrial sludge 100t/ha mud (table 3). However, this decrease was significantly marked with the addition of urban sludge in the siliques and leaves. The Mn content in different parts of the plant decreased with increasing dose of mud. Indeed essential divalent cations such as  $Mn^{2+}$ ,  $Fe^{2+}$  are competitors vis-a-vis the toxic metals (Cataldo et al., 1983; Costa & Morel, 1994).



Figure 12. Distribution of Co contents in Canola plants grown in the different dose of urban and industrial sludge. Témoin: control soil.



Figure 13. Distribution of Cu contents in Canola plants grown in the different dose of urban and industrial sludge.

#### 4. DISCUSSIONS AND CONCLUSION

Considering their composition of organic matter and nutrients, sludge is of suitable use in agriculture or to rehabilitate degraded agricultural land. In this same section, showed that the instantaneous fertility and soil physical properties are improved by the application of sludge.

In this study, several analyses were realised to collect information about the texture, mineral and crystalline composition of the sludge. A significant difference was observed between the chemical composition of urban sludge and the industrial sludge with a dominance of all trace metals including Cd, Pb and Cr. The use of X-rays to determine the crystal structure showed that the sludge consists mainly of quartz, gypsum and calcite. The daubreelita, brezininaite, wattersite are present in industrial sludge explaining its high Cr concentration. The exact composition of the sludge depends on the origin of the waste water, the year period and the type of processing and packaging made in the wastewater treatment station (Werther & Ogada, 1999; Jarde et al., 2003; Singh et al., 2004).

Sludge improves the production of dry biomass of Canola and leaf area including mature leaves, but this improvement is more important for urban sludge. The improvement in grain yield was due to an increase in the number of seed and not improved seed weight. This improvement could be related to a continuous release of N that feeds the. Similar results were obtained by Gukert & Morel (1979) who observed a significant ameliorative action of sludge with or without mineral fertilization on the production of English Ray Grass compared to a control without sludge. Work on the Ray Grass made by Nejmeddine et al. (2003) also showed a beneficial effect of sewage sludge. According to Singh & Sinha (2005), this beneficial effect was attributed to the effect of sludge on soil fertility by improving the availability of certain nutrients such as nitrogen, phosphorus, potassium, magnesium and by holding capacity of the soil.

The many authors (Guckert In rerefences lis is Gukert & Morel, 1979; Larry, 1981; Rejeb & Bahri, 1995; Rejeb et al, 2003; Zaier et al., 2010; Roberts et al., 1988 and Pigozzo et al., 2000) were reported a significant increase in yields of various crops such as sorghum, corn, chili, potato, ryegrass and fescue.

If it is proved that the application of sludge promotes the productivity of crops, it is nevertheless accompanied by an accumulation of heavy metals (Boukhars & Rada, 2000; Big et al., 2012; Miller et al., 1995; Juste et al., 1995; Nicholson et al., 2003); Galfati et al., (2011); Damian et al. (2008). Work done by Juste & Mench (1992) showed that most of the metals accumulate in the surface soil horizon which presents a limit to this practice (Nyamangara & Mzezewa, 1999). Depending on the plant and the element, the sample can be located either at the apical root region, the entire root surface, but very little data exist (Wierzbicka, 1998).

The results of this study showed that the addition of sludge increased the levels of Cd, Pb, Cr, Ni and Zn in plant especially in the roots. This increase is dependent on the dose of sludge and especially on the types of sludge with its different quantity of these elements. The increase of these elements in the plant in the presence of sludge had often been found by several authors (Garcia-Hernandez et al., 1998, Rejeb, 1990, Rejeb et al., 2011). The high mobility of Cd and Zn could explain the migration of these elements in the plant and especially the accumulation of Cd observed in the seeds of 100t/ha of industrial sludge treatment. This could be the result of an overflow of capacity control of the root system (Rejeb, 1999). It also seems that the type of sandy soil of the test plot which has a coarse texture and CEC content of amorphous iron oxide low 0.95% could favor increasing Cd and Zn in the plant (Chang et al., 1983 and Kuo et al., 1985).

The most important changes of the Pb were fixed at the roots which also had the highest values compared to other parts of the plant. The Pb appeared as one of element the most used by the roots that played the role of a barrier against the migration to the aerial part of plants (Rejeb, 1990). The roots of Canola also retained the Cr which has the property of binding to the cell walls of the root and does not migrate into the aerial part. The attachment of the Pb and Cr in the roots helps reduce phytotoxic effects.

Fe contents and to a less extent Mn contents had been reduced as a result of sludge despite the richness of the two types of sludge in these elements. These results were consistent with those obtained by Morel et al., (1988) and Juste & Solda (1977). The interaction with one or more elements present in significant amounts in the environment, especially Zn may explain the decrease of Fe. For manganese, Rejeb (1990, 1999) attributed the variability in that element to the more or less iron presence in the sludge. The fall of manganese could be due to a dilution effect caused by an increase in biomass or soil pH (Soon et al., 1980; Morel, 1985; Morel et al., 1988).

It should be noted that in general, the concentrations of Cd, Co, Cr, Pb, and Ni were lowered in seeds compared to other organs.

#### ACKNOWLEDGEMENTS

This work was supported by National Institute for Rural Engineering research, Water and Forestry, Tunis

and Geosciences & Environment Department of Ecole Nationale Supérieure des Mines de Saint Etienne, France. We thank prof. Dr. Peter Andráš for his remarks and suggestions to improve the writing of the paper.

#### REFERENCES

- Abrahams, P.W., 2002. Soils: their implications to human health. The Science of the Total Environment, 291, 1-32.
- Adriano, D.C., 2001. Trace elements in terrestrial environments: biogeochemistry, bioavailability and risks of metal. 2nd Springer-Verlag, New York, Berlin, Heidelberg, 866 p.
- Aldrich, M.V., Gardea-Torresdey, J.L., Peralta-Videa. & J.R., Parsons, J.G., 2003. Uptake and reduction of Cr(VI) to Cr(III) by mesquite (Prospis spp.): chromate-plant interaction in hydroponics and solid media studied using XAS. Environ. Sci. Technol, 37, 1859–1864.
- Barbe, J., Brocheton, D., Kockmann, F. & Wiart, J., 2002. Limed sludge from municipal sewage production, quality and agronomic stations. ADEME, 7 p.
- Ben Ghnaya, A., Gilbert, Ch., Hourmant, A., Ben Hamida, J. & Branchard, M., 2009. Physiological behaviour of four rapeseed cultivar (Brassica napus L.) submitted to metal stress. Elsevier Masson SAS on behalf of Académie des sciences, 332, 363–370.
- Big, C.A., Lăcătşu, R. & Damian, F., 2012. *Heavy* metals in soil-plant system around Baia Mare city, *Romania*. Carpathian Journal of Earth and Environmental Sciences, 7(3), 219-230.
- Boukhars, L. & Rada, A., 2000. Plant exposure to cadmium in Moroccan calcareous salty soils treated with sewage sludges and wastewaters. Environ. Technol, 21, 641-652.
- Cataldo, D., McFadden, K.M., Garland, T.R. & Wildung, R.E., 1983. Organic constituents and complexation of nickel (II), iron (III), cadmium (II), and plutonium (IV) in soybean xylem exudates. Plant Physiology, 86 p.
- **Coullery, P.,** 1997. Behavior of heavy metals in temperate agro low pollution levels. Ecole Polytechnique Federale de Lausanne, 138 p.
- **Costa, G. & Morel, J.L**., 1994. *Efficiency of H+-ATPase activity on cadmium uptake by four cultivars of lettuce.* J. Plant Nutrition, 14, 627-637.
- Chang, A.C., Page, A.L., Warneke, J.E., Resketo, M.R. & Jones, T.E., 1983. Accumulation of cadmium and Zn in barley grown on sludgetreated soils: a longterm field study. J. Envir. Qual,12, 391-397.
- Damian, F., Damian, Gh., Lăcătuşu, R., Macovei, Gh., Iepure, Gh., Năprădean, I., Chira, R., Kollar, L., Rață, L. & Zaharia, D,C., 2008. Soils from the Baia Mare zone and the heavy metals pollution. Carpthian Journal of Earth and Environmental Sciences, 3(1), 85-98.

- Galfati, I., Bilal, E., Beji Sassi, A., Abdallah, H., & Zaier, A., 2011. Accumulation of heavy metals in native plants growing near the phosphate treatment industry, Tunisia. Carpathian Journal of Earth and Environ-mental Sciences, 6(2), 85–100.
- Garcia-Hernandez, M., Murphy, A. & Taiz, L., 1998. Metallothioneins 1 and 2 have distinct but overlapping expression patterns in Arabidopsis. Plant Physiology, 118, 387–397.
- Gukert, A. & Morel, J.L., 1979. Taking stock of five years of use of urban sewage sludge on field crops in Lorraine agro-climatic conditions. First European symposium of treatment and use of sewage sludge, Ed Alexendre, 269-282.
- Jarde, E., Mansuy, L. & Faure, P., 2003. Characterization of the macromolecular organic content of sewage sludges by thermally assisted hydrolysis and methylation-gas chromatographymass spectrometer (THM-GC/MS). J. Anal. Appl. Pyrol, 69, 31-350.
- Juste, C., Chassin, P., Gomez, A., Linères, M., Mocquot, B., Feix, I. & Wiart, J., 1995. The metal micronutrients in sewage sludge from urban waste water treatment plants. Ademe, 209 p.
- Juste, C. & Mench, M., 1992, Long term leaching of trace elements in heavy sludge-amended silt clay lam soil. Soil Sci., 164(9), 613-623.
- Juste, C. & Solda, P., 1977. Effect of application of sludge urban wastewater treatment plants in monoculture corn. Effect on yield and composition of plants and some soil characteristics, soil science, 3, 147-155.
- Kabata-Pendias A. & Pendias H., 1992. *Trace elements in soils and plants*. Published by CRC Press, Boca Raton, USA, 365 p.
- Kalra, Y.P. & Maynard, D.G., 1991. Methods for Forest Soil and Plant Analysis. Information Report NOR-X-319, Forestry Canada, Northwest Region, Northern Forestry Center, 116.
- Kuboi T., Noguchi A. & Yazaki J., 1986. Familydependent cadmium accumulation characteristics in higher plants. Plant Soil, 92, 405-415.
- Kuo, S., Jellum, E.J. & Baker, A.S., 1985. Effects of soil type, liming, and sludge application on Zn and Cd availability to Swiss chard. Soil Science, 139, 122-130.
- Lăcătuşu R., Cîtu G., Aston J., Lungu M. & Lăcătuşu A.R., 2009. Heavy metals soil pollution state in relation to potential future mining activities in the Roşia Montană area. Carpathian Journal of Earth and Environmental Sciences, 4(2), 39-50.
- Larry, D.K., 1981. Effects of swine manure lagoon sludge on grawth, a review of plant, soil, metal chemical interaction to increase the bioavailability of trace metals in the ground, Agronomy, 16, 201-215.
- McGrath, S.P. & Cunliffe, C.H., 1985, A simplified method for the extraction of the metals Fe, Zn, Cu, Ni, Pb, Cr, Co and Mn from soils and sewage sludges, J. Sci. Food Agriculture, 36, 794–798.

- Mench, M. & D. Baize., 2004, Contamination of soil and our plant foods by trace elements, measures to reduce exposure, Courier environment INRA, 52, 31-56.
- Miller, R.W., Azzari, A.S. & Gardiner, D.T., 1995, Heavy metals in crops as affected by soil types and sewage sludge rates, Plant Anal., 26(5-6), 703-711.
- Morel, J.L., Pierrat, J.C, & Guckert, A., (1988), Study and effect back spreading urban sludge coordinates with lime and ferric chloride on the heavy metal content of corn, Agronomy, V. 8, 107-113.
- Nejmeddine A., Echab A., Fars S. & Hafidi M., 2003, Accumulation of trace elements by ryegrass grown on soils amended by sewage sludge, Cahiers Agricultures V. 12, 33-38.
- Nicholson, F.A., Smith, S.R., Alloway, B.J., Carlton-Smith, C. & Chambers, B.J., 2003, An inventory of heavy metals inputs to agricultural soils in England and Wales, Science of the Total Environment, 311, 205-219.
- Nyamangara, J. & Mzezewa, J., 1999, The effect of long-term sewage sludge application on Zn, Cu, Ni and Pb levels in a clay loam soil under pasture grass in Zimbabwe, Agric. Ecosyst. Environ, 73, 199–204.
- Petrescu L. & Bilal E., 2003. Plant availability of uranium in contaminated soil from Crucea mine (Romania). Environmental Geosciences, 10(3), 123-135.
- Petrescu, L., & Bilal, E, 2006. Natural actinides studies in conifers grown on uranium mining dumps (The East Carpathians, Romania), Carpthian Journal of earth and environmental sciences, 1(1), 63–80.
- Petrescu, L. & Bilal, E., 2007. Environmental impact assessment of a uranium mine, east Carpathians, Romania: metal distribution and partitioning of U and Th, Carpathian Journal of Earth and Environmental Sciences, 2(1), 39–50.
- Pigozzo, A.T.J., Gobbi, M.A., Lenzi, E. & Luchese, E.B., 2000. Effects of the application of sewage sludge and petrochemical residue in maize culture as source of micronutrients on soils of Parana state. Braz. Arch. Biol.Technol, 43, 143–149.
- **Rejeb, S. & Bahri,** A., 1995. Impact of the contribution of urban sewage sludge on the mineral composition and productivity of some crops grown in Tunisia, Books of the RMAF, 24, 13–31.
- Rejeb, S., Gharbi, F., Mouneimne, A.H. & Ghorbal, M.H., 2011. Effects of sludge on the metals levels of various edible crops grown in the field, ARPN Journal of Agricultural and Biological Science, 6, 1990-6145.
- Rejeb, S, Khelil, M.N., Gharbi, F. & Ghorbal, M.H., 2003. *Effect of sewage sludge on potato*, Libbey Eurotext, Montrouge, FRANCE, 12, 39-42.

- Roberts, J.A., Daniels, W.L., Bell, J.C. & Martens, D.C., 1988. Tall fescue production and nutrient status on southwest Virginia mine soils. J. Environ. Qual, 17, 55–61.
- Savva, A.P. & Karen, F., 2002. Planning, Development Monitoring and Evaluation of Irrigated Agriculture with Farmer Participation, Food and Agriculture Organization of the United Nations (FAO) Sub-Regional Office for East and Southern Africa (SAFR), V. 5. pp. 149.
- Secu, C.V., Iancu, O.G. & Buzgar, N., 2008. Lead, zinc and copper in the bioacumulative horizon of soils from Iaşi and the surrounding areas, Carpathian Journal of Earth and Environmental Sciences, 3(2), 131-144.
- Sellami, R., Gharbi, F., Rejeb, S., Rejeb, M.N., Henchi, B., Echevarria, G. & Morel, J.L., 2012. Effects of Nickel Hyperaccumulation on Physiological Characteristics of Alyssum Murale Grown on Metal Contaminated Waste Amended Soil, International Journal of Phytoremediation, 14(6), 609-620.
- Singh, K.P., Mohan, D., Sinha, S. & Dalwani, R., 2004. Impact assessment of treated/untreated wastewater toxicants discharged by sewage treatment plants on health, agricultural, and environmental quality in the wastewater disposal area. Chemosphere, 55. 227–255.
- Singh, S. & Sinha, S., 2005. Accumulation of metals and its effects in Brassica juncea (L.) Czern. (cv. Rohini) grown on various amendments of tannery waste. Environ. Safety, 62, 118–127.
- Soon, T.K., Bates, T.E., Beauchamps, E.G. & Moyer J.R., 1980. Land application of chemically treated sewage sludge: effect on soil and plant, Environ Qual, 9, 497-504.
- Veerle Grispen, M.J., Hans Nelissen, J.M. & Jos Verkleij A.C., 2006. Phytoextraction with Brassica napus L. A tool for sustainable management of heavy metal contaminated soil, Environ. Pollut, 144, 77–83.
- Werther, J. & Ogada, T., 1999. Sewage sludge combustion, Progress in Energy and Combustion Science, 25, 55–116.
- Wierzbicka, M., 1998. Lead in the apoplast of Allium cepa L. root tips ultrastructural studies, Plant Science, 133, 105-119.
- Zaier, H., Ghnaya, T., Ben Rejeb, K., Lakhdar, A., Rejeb, S. & Jemal, F., 2010. Effects of EDTA on phytoextraction of heavy metals (Zn, Mn and Pb) from sludge-amended soil with Brassica napus, Bioresource, Technology, 101, 3978–3983.
- Zarcinas, B.A., Cartwright, B. & Spouncer, L.R., 1987. Nitric acid digestion and multielement analysis of plant material by inductively coupled plasma spectrometry. Comm Soil Sci Plant Anal 18, 131-146.

Received at: