Evaluation of the application potential of disinfected biosolids on radish crop

Evaluación del potencial de aplicación de biosólidos higienizados en el cultivo de rábano

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Abstract

We evaluated the use potential in the cultivation of radish of three types of biosolids: dehydrated- BD, thermally dried (60 °C for 12.58 hours)-BST and alkalinized (quicklime 9% dry weight)-BA in doses of 1, 2, 4 and 8 times the nitrogen requirement of the crop. We measure the number of leaves, biomass and weight of the radish and the content of fecal coliforms, *E. coli* and helminth eggs in the radish at the moment of harvest. The results showed that application of biosolids improves the weight of the radish from 17 to 150% in relation to the control, however, the statistical analysis showed no significant difference between treatments. Although the application of biosolids did not show a concentration of fecal coliforms and *E. coli* in radish for the three biosolids; the dehydrated biosolid application increased the content of viable helminth eggs in radish, presenting an elevated risk for use on this crop, being necessary to implement sanitation practices of biosolids before the agricultural application

Key words: Agricultural application, alkaline treatment, biosolids, radish, thermal drying.

Resumen

Se evaluó el posible aprovechamiento de tres tipos de biosólidos, procedentes del manejo de los lodos de plantas de tratamiento de aguas residuales domésticas, para el cultivo y producción de rábano (Raphanus sativa) cultivado en un suelo Vertic endoaquepts. Los tratamientos de los biosólidos consistieron en: deshidratado-BD, secado térmicamente (60 °C por 12.58 h)-BST y alcalinizado (cal viva 9% peso seco)-BA, aplicados en dosis de 1, 2, 4 y 8 veces el requerimiento de nitrógeno del cultivo (80 kg/ha). En el cultivo se midieron el número de hojas de las plantas y la producción de biomasa y en los rábanos el peso final y el contenido de coliformes fecales, *Escherichia coli*, y huevos de helmintos al momento de la cosecha. Los resultados mostraron que la aplicación de biosólidos mejoró entre 17% y 150% los pesos de la planta y del rábano en relación con el tratamiento control; no obstante, no se encontraron diferencias ($P > 0.05$) en el peso del rábano entre tratamientos. Aunque la aplicación de los biosólidos no influyó en la concentración de coliformes fecales ni de *E. coli* en el rábano al momento de la cosecha, la aplicación de estos aumentó el contenido de huevos de helmintos viables, lo que presenta un riesgo elevado para la salud humana, y hace necesario implementar prácticas de higienización antes de su uso en cultivos.

Palabras clave: Aprovechamiento agrícola, biosólidos, rábano, secado térmico, tratamiento alcalino.

Introduction

Biosolids coming from sludge treatments of domestic wastewater treatment facilities – WWTPs have a high potential to be used in agriculture due to its organic matter and nutrient content. However, the presence of pathogenic microorganisms is a constrain for its direct use without a previous treatment, because it is a threat to human and biotic health. Therefore, it is needed the implementation of complementary treatments for stabilization such as thermic dry and alkaline treatments, among others (Kiely, 1999; EPA, 1999).

Thermic dry allows effective control of temperature and exposition time and has a high potential for sanitation as complement to its modest efficiency in pathogen removal from the biological digestion of sewage sludge (Almeida *et al.*, 2006). It has been found that with temperatures higher than 45 °C is possible to significantly eliminate spore forming bacteria like *Clostridium perfringens, Bacillus* spp. and sulfite reducing, additionally to eliminate highly persistent and pathogenic microorganisms like helminth eggs (Gantzer, 2001). Recommended temperature and time ranges vary between 50 and 75 °C and between 3 min and 13 days, which depends on the type of microorganism to eliminate (Comparini and Sobrinho, 2002; Franco-Hernández *et al.*, 2003; Bagge *et al.*, 2005; Araque, 2006; Pecson *et al.*, 2007; Lang and Stephen, 2008; Avery *et al.*, 2009; Sidhu and Toze, 2009).

Alkaline treatment is one of the most used to sanitize biosolids and consists in alkaline stabilization using a base, normally lime, which is mixed with the biosolid to increase pH and destroy most of pathogenic microorganisms (Fernández and de Souza, 2001). According to EPA (2003) this process is reached by increasing the pH >12 during 72 h or mores and temperature at 52 °C for 12 h period.

Biosolids application in the soil is frequent in forestry (forest or nursery), to recover degraded soils, to adequate green zones, fertilizer or organic amendment and, for bioremediation of contaminated soils (Castrejón *et al.*, 2000). Its potential for use relies on the previous treatment and type of crop where it will be applied. Crops on top of the soil and for direct consumption without previous treatment have the widest restrictions because of the risk of pathogen presence.

Nitrogen volatilization is the most significant lost while the biosolids incorporation as soil fertilizer. This process depends on temperature, soil pH, cationic exchange capacity, organic matter content, cover and quality of superficial residues, wind, superficial water pressure and applied doses (Sogaard *et al.*, 2002; Ferraris *et al.*, 2009).

Hoitink *et al.* (2003) observed that the application of community biosolids without sanitation at rates of 5.6% in volume helped the growth of 40 plant species, among them red radish (*Raphanus sativa*); to the contrary, Evanylo *et al.* (2005) and Lopes (2008) did not find changes in radish development and production with the application of different doses of biosolids to the soil.

In Colombia there are several studies about application of muds and biosolids in crops. Ramírez and Pérez (2006) evaluated the agricultural potential of biosolids of El Salitre WWTP in red radish in variable doses between 25% till 100% of the nitrogen requirements and found that in doses between 25% and 50% growth and development of the crop was increased. Ramírez *et* *al.* (2007) observed that the application of biosolids of San Fernando WWTP in Medellin, that have 4% and 8% of OM negatively affected the growth of gualanday (*Jacaranda mimosifolia*) due to the high content of nutrients and heavy metals in the soil. Torres *et al.* (2007) evaluated the agricultural application of compost from primary biosolids coming from the Cañaveralejo WWTP in Cali, in radish (*Raphanus sativus*) and chard (*Beta vulgaris*) and did not find differences in relation to the application of commercial fertilizer 10-30-10; whereas in chard there were increments between 43% and 130% in comparison to the control soil. In Santander, Escobar (2010) evaluated the effect of application of biosolids from Rio Frio WWTP, which is stabilized with agricultural lime on a dose of 33% in lecttuce (*Lactuca sativa*), guadua (*Guadua* spp), saman (*Samanea saman*) and found a positive effect mainly in lettuce plants.

This work has as objective to evaluate the potential of use of dehydrated biosolids of the Cañaveralejo wastewater treatment plant, PTAR-C, Cali using radish as indicator crop.

Materials and methods

This work was performed on a Vertic endoaquepts soil (USDA, 1999) which is representative of 9777 ha of the Cauca river geographic valley (Carbonell *et al.*, 2006). Biosolids were taken from the PTAR-C that operates with the advanced primary treatment (APT) where sludges are led for thickening treatments, anaerobic digestion and mechanical dehydration by filter press – BD; additionally, it was evaluated biosolids previously subjected to pathogen reducing processes by thermal drying at 60 °C for 12.58 h (BDST) and alkaline treatment with 9% lime dry weight (BDTA), according to recommendation by Silva *et al.,* (2013).

Biosolids were characterized by their physical and chemical aspects such as pH, humidity, organic content (Walkley and Black, 1934), total nitrogen (Kjeldahl, 1883), ammonia nitrogen, nitrates, nitrites, phosphorus (Bray y Kurtz, 1945), potassium, calcium, magnesium, sulfur and heavy metals (As, B, Cd, Mn, Fe, Cu, Zn, Hg, Ni, Ag, Pb and Cr) (USDA and NRCS, 2004); microbiological aspects like total and fecal coliforms and *E. coli* (APHA *et al.*, 2005); and parasitology with helminth eggs (Bailinger, 1979).

Treatment with biosolid was applied to the soil to supply the nitrogen requirement for radish, which is equivalent to 80 kg/ha (CHC, 2012) based on the Equation 1 and the available nitrogen was calculated based on the Equation 2 (CETESB, 1999).

The constant of mineralization of each biosolid (Kmin) was determined by the method proposed by Silva *et al.* (2013). Applied treatments were equivalent to 1, 2,

$$Biosolid\ dose\ = \frac{kg}{ha} = \frac{Recomendend\ nitrogen\ for\ the\ crop\ \frac{kg}{ha}}{Available\ nitrogen\ in\ the\ biosolid\ \frac{mg}{kg}\ x\ 0.001} \quad Eq. 1$$

$$ND = \frac{Kmin}{100} x\ (NTK\ \ NH_4^+) +\ 0.5\ (NH_4^+) +\ (NO_3 +\ NO_2) \quad Eq. 2$$

where:

ND	available nitrogen (mg/kg),
Kmin	constant of mineralization of each biosolid, Kjeldahl (mg/kg),
NH_4^+	ammonia nitrogen in the biosolid (mg/kg),
NO_3^- and NO_2^-	nitrates and nitrites in the biosolid,
0.5	volatilization factor of ammonia nitrogen by superficial aplication,
0.001	conversion factor from mg/kg to kg/t.

4 and 8 times the crop requirements (Table 1) proposed by Vieria and Cardosos (2003), Vieira (2004), Vieira *et al.* (2005), Guimarães (2007) and Chiba *et al.* (2008 a,b).

As indicator crop it was used red radish of fast germination (7 days) and short life cycle (45 days to harvest) (CHC, 2012). Each experimental unit was composed of 0.7 l pots with 4 seeds per pot. The experimental design was factorial with two factors: biosolid type (dehydrated, thermal and alkaline) and applied doses, for a total of 13 treatments including the control treatment, with two replicates for a total of 26 experimental units. At harvest, 45 days after sowing, number of leaves, plant aerial biomass and radish fresh weight were measured. In these, a microbiological and parasitological analysis was performed to determine the content of fecal coliforms, *E. coli* and helminth eggs by the modified Bailinger method (1979) (Table 2). The statistical analysis was done by Anova using the statistical software R.2.10.

Results and discussion

Biosolid characterization

Soil analysis showed that the organic carbon content was low (Table 3), according to

Table 1. Types of biosolids and doses applied to radish.

Treatment (type of biosolid)	**Equivalent requirement for radish**	**K mineralization (%)**	**Amount applied (t/ha)**
Control (soil)	0		0
Dehydrated -BD	1	33	9.3
	2		18.6
	4		37.2
	8		74.4
Thermally dried	1	45.7	6.7
-BST	2		13.5
	4		26.9
	8		53.9
Alkalinized -BA	1	26	16.8
	2		33.7
	4		67.5
	8		134.9

Table 2. Measured variables in the crop and frequency.

Variable	**Unit**	**Frequency**	**Method**
Leaf number	Number	End of the crop	Direct
Plant biomass	Grams	End of the crop	Gravimetric
Radish weight	Grams	End of the crop	Gravimetric
Fecal colliforms	CFU/g	End of the crop	Membrane filtration
E. coli	CFU/g	End of the crop	Membrane filtration
Helminth eggs	HH/g	End of the crop	Bailinger, 1979 modified

Table 3. Initial characterization of soil and dehydrated, thermally dried and alkaline biosolids

Variable	Soil	Type of biosolid			Reference value
		Dehydrated (BD)	Thermically dried(BDST)	Alkaline (BDA)	
pH	7.4	7.7	7.8	12.1	5.5-6.4[a]
Moisture (%)	27.4	67.4	16.8	57.9	71.2-82.7[a]
Organic carbon (g/kg)	6.8	243.1	257.4	218.2	201-382[a]
N-Total, Kjeldahl (g/kg)	–	25.0	25.8	18.0	21.0-68.2[b]
$N\text{-}NH_4$ (mg/kg)	8.1	1824.7	1130.7	113.1	1403-10253[b]
$N\text{-}NO_2$ (mg/kg)	1.7	0.0	0	0	–
$N\text{-}NO_3$(mg/kg)	4.4	33.8	17.8	34.5	51-106[c]
P-Total (mg/kg)	7.6	14.5	14.3	9.8	0.95-26.9[a]
K (g/kg)	0.2	1.0	0.94	0.72	0.34-1.0[a]
Ca (g/kg)	21.7	35.4	31.9	137.5	6.6-47.8[a]
Mg (g/kg)	9.0	5.5	5.7	5.2	1.3-6.0[a]
S-Total (g/kg)	27.4	17.6	13.8	11.5	–
Arsenic (ppm)	0.17	0.64	0.53	0.58	<1
Borum (ppm)	0.82	0.93	0.93	0.92	10.1-29.3
Cadmium (ppm)	<0.37	0.65	0.77	0.77	2.1-9.4
Manganese (mg/kg)	21.1	386.2	378.2	270.1	232-425 1
Iron (mg/kg)	0.2	36434.6	37168.5	34127.3	15730-37990
Copper (ppm)	63	96.98	126.63	141.82	114-953
Zinc (mg/kg)	0.4	1068.3	1163.1	861.2	530-3372
Mercury (ppm)	<0.025	1.07	0.77	0.72	0.01-1.8
Níckel (ppm)	44.26	42.84	48.57	57.21	25.3-605.8
Silver (ppm)	<0.50	3.8	3.32	2.95	–
Lead (ppm)	3.58	28.07	34.9	31.55	60-349
Chrome (ppm)	65.75	43.99	63.03	62.58	53-1230
total colliforms/g	1.0×10^5	$1.94 \times 10^{7*}$	$1. \times 10^2$	0	–
Fecal collliforms UFC/g	0	$2.08 \times 10^{6*}$	0	0	$<1 \times 10^3$[c]
E. Coli CUF/g	0	$1.70 \times 10^{6*}$	0	0	–
Helminth eggs no./g	0	9	0	0	<1[d]

[a.]Bettiol and Camargo (2008), [b.]Dynia *et al.* (2008), [c.]Vieira *et al.* (2005), dEPA (2003) class A.

the categories defined by Quintero (1993) for the soils of the geographic valley of the Cauca river. The chemical composition of the dehydrated biosolids was similar to the one found by Vieira *et al.* (2005), Bettiol and amargo (2008) and Dynia *et al.* (2008) for the biosolids from waterwaste treatment plants in Brazil, which normally use aerobic treatment systems in ponds and aeration tanks and the sludge is dehydrated and digested by anaerobic ways. Composition of thermally dried biosolid was similar to the results of Ahmith and Durham (2002), Tarrasón *et al.* (2008) and Ramírez *et al.* (2008) who did not observe any difference between thermally dried and dehydrated biosolids. The alkaline treatment reduced the organic carbon and ammonia nitrogen of the biosolid, indicating that this treatment significantly affects these properties and confirm the findings of Czechowski and Marcinkowski (2006) who found nitrogen losses of 28% in comparison to dehydrated biosolids.

The evaluated biosolids did not show limitations for the heavy metals content, according to the EPA (2003) recommendations; however, because of its pathogen and parasite content, the dehydrated biosolid was similar to the one obtained by Guzmán and Campos (2004), Ramírez and Pérez (2006), Araque (2006) and Torres *et al.* (2009), this material has restrictions for its use since it does not accomplish the EPA requirements for class A and B biosolids.

The thermal drying and alkaline treatment guarantee obtaining a class A biosolid that can be used in agriculture without restrictions about microbes.

Plant characteristics

The application of different biosolids doses did not affected the number of leaves, neither the crop development when compared with the control treatment (Figure 1). These results are similar to the ones obtained by Ramírez and Pérez (2006) and Lopes (2008) who found that biosolid applications in doses up to 294 t/ha did not affect the development of red radish crop.

In Figure 2 is observed the beneficial effect of the biosolid incorporation in the soil on the total biomass increase in radish plants. Excepting the control treatments, the biomass weight of the plants was similar to the other treatments and varied between 8.8 and 11.6 g.

The fresh weight of the radishes (Figure 3) and the plant biomass, was positively affected by biosolids application, however, in this case differences were observed between types, showing less effect the alkaline ones and higher effect the ones thermally dried. This difference is due to the fact that the thermal drying process increases the mineralized nitrogen content in the biosolid, making them an effective and fast nitrogen source for the plants (Rigby *et al.*, 2009). The statistical analysis of radish weight showed that there is no significant difference between biosolids compared to the control (P = 0.249). Nevertheless the weight increased between 40% and 150% for the radishes in the dehydrated biosolid, between 92% and 109% in the thermally dried biosolid and, between 17% and 40% in the alkaline biosolid.

Pathogen presence

Results in Figure 4 show that biosolid application did not affect the abundance of fecal coliforms and *E. coli* in radish at the end of the crop. For the dehydrated biosolids helmint eggs were found in the radish, they increase as the applied dose was increased; this raises a risk on human health due to the use of this biosolid which was demonstrated by Gerba (1983) who found that some parasites can survive for months or years, which depends on the weather and soil conditions.

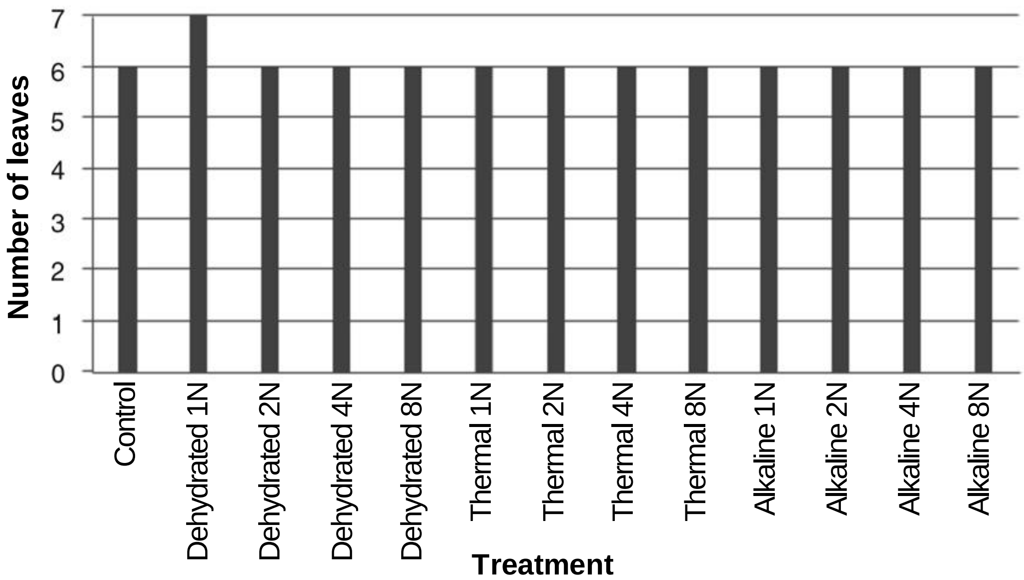

Figure 1. Number of final leaves in each applied treatment. 1N: Once the nitrogen requirement of the crop; 2N: Twice the nitrogen requirement of the crop; 4N: Four times the nitrogen requirement of the crop; 8N: Eight times the nitrogen requirement of the crop.

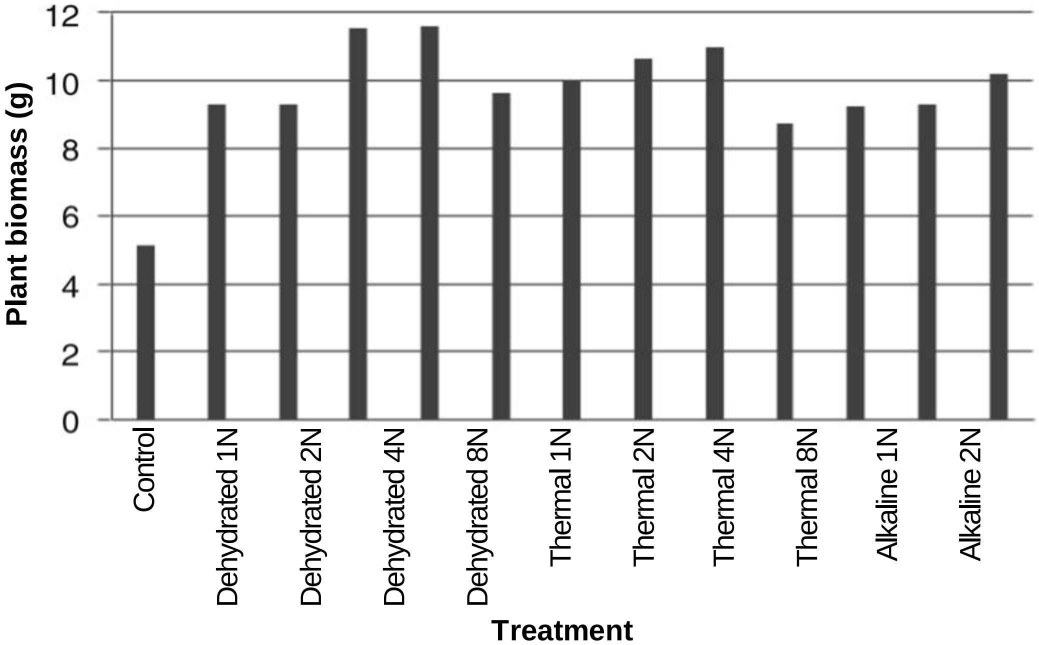

Figure 2. Total biomass of radish plants. 1N: Once the nitrogen requirement of the crop; 2N: Twice the nitrogen requirement of the crop; 4N: Four times the nitrogen requirement of the crop; 8N: Eight times the nitrogen requirement of the crop.

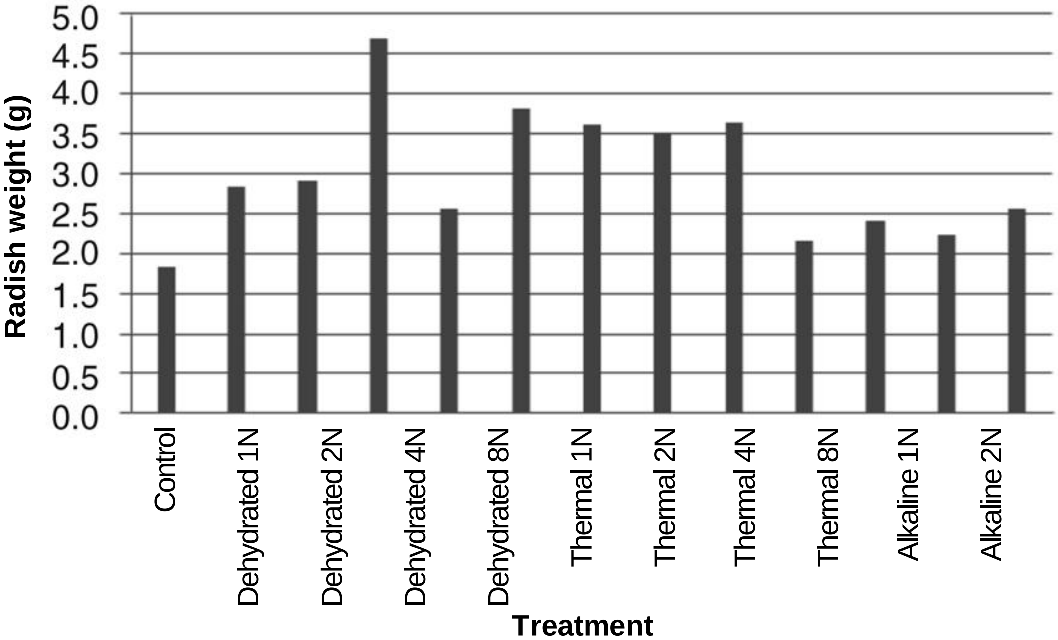

Figure 3. Radish weight at harvest. 1N: Once the nitrogen requirement of the crop; 2N: Twice the nitrogen requirement of the crop; 4N: Four times the nitrogen requirement of the crop; 8N: Eight times the nitrogen requirement of the crop.

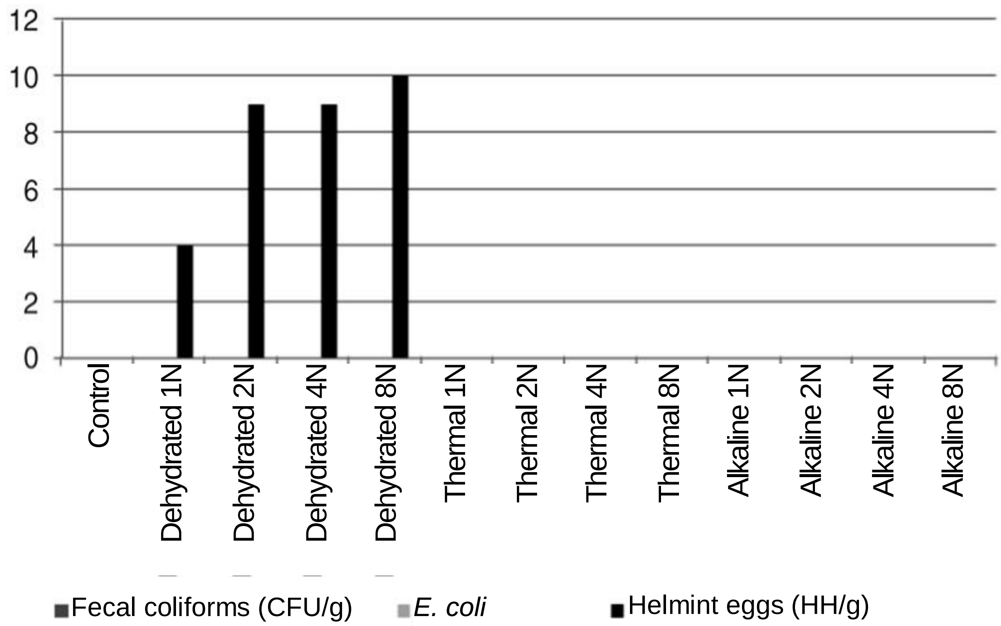

Figure 4. Microbiology results of the radish at the end of the crop. 1N: Once the nitrogen requirement of the crop; 2N: Twice the nitrogen requirement of the crop; 4N: Four times the nitrogen requirement of the crop; 8N: Eight times the nitrogen requirement of the crop

Conclusions

- Biosolids produced at the waterwaster treatment plant in Cañaveralejo, Cali, Colombia, did not show physic-chemical or microbiological restrictions for their use in agriculture, except the dehydrated biosolid that had values of 2.08 x 10^6 CFU/g for fecal coliforms and 9 HH/g helminth eggs that limit its use in agriculture.
- Biosolids application in the soil improved up to 150% the plant aerial part and radish weight in relation to the control treatment.
- Dehydrated biosolid application increased the content of helminth eggs in the radish, which is a high risk for human health and confirms the need to use sanitary treatments in the biosolids prior their use in agriculture.

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