Divergency of compost extract and organic manure effects on lucerne plant and soil

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Aim: Application of organic materials into agricultural systems enhances plant growth, yields, and improves soil fertility and structure. This study was undertaken to examine the effects of “compost extract (CE)”, a soil conditioner, and organic manure (OM) on the growth of lucerne (*Medicago sativa*), and compare the efficiency between OM, which included numbers of microorganisms and CE, which included no added microorganisms. Method: A greenhouse experiment was conducted with four soil amendment treatments (control, OM, CE and CE + OM) , and was arranged in a completely randomized design with 10 replicates. Plant biomass, nutritive value and rhizobia efficacy as well as soil characteristics were examined. Result: CE rather than OM application showed a positive effect on plant growth and soil properties when compared with the control. Lucerne nodulation responded equally between CE adding and rhizobium inoculating. CE alone and in combination with OM significantly increased plant growth and soil microbial activities and improved soil structure in the study location. The synergistic effects of CE and OM indicate that applying CE and OM together could increase their efficiency, leading to higher economic returns and improved soil health. However, CE alone is more effective for legume growth since nodulation was suppressed by external nitrogen from OM. CE had a higher efficiency than OM for enriching the indigenous microorganisms in the soil instead of adding additional microorganisms and plant nodulating.

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Running title: Compost extract improve lucerne nodulation process

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Abstract

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Result: CE rather than OM application showed a positive effect on plant growth and soil properties when compared with the control. Lucerne nodulation responded equally between CE adding and rhizobium inoculating. CE alone and in combination with OM significantly increased plant growth and soil microbial activities and improved soil structure in the study location. The synergistic effects of CE and OM indicate that applying CE and OM together could increase their efficiency, leading to higher economic returns and improved soil health. However, CE alone is more effective for legume growth since nodulation was suppressed by external nitrogen from OM. CE had a higher efficiency than OM for enriching the indigenous microorganisms in the soil instead of adding additional microorganisms and plant nodulating.

Keywords: compost extract, organic manure, soil amendment, indigenous microorganism, nodulation

Introduction

Preserving and restoring native grasslands and agricultural production systems by improving soil quality have been common goals worldwide (Henwood 2010). However, long-term inappropriate management and large inputs of chemical fertilizers and pesticides into soils have led to severe soil problems and thus emerged types of soil conditioners. These commercial soil conditioners or manures are used in agricultural production systems for amendment of nutrient-deficient soils by providing multiple nutrients (Soumare et al. 2003, Hu and Qi 2013), including but not limiting to organic manure (OM), effective microorganisms (EM), and compost tea. However, farmers are sometimes reluctant to use organic fertilizers because the effects of organic nutrients on plant growth are not as quickly seen as inorganic fertilizers. Thus, new materials and new techniques for increasing the efficiency of organic materials are needed.

Introducing exogenous microorganisms into environments has been applied to accelerate bioremediation and improve agricultural productivity (Paluch et al. 2013). Although less is known about the practical effectiveness and mechanisms of these products, assessment of their impacts have focused on their potential to increase the soil organic matter level, promote nutrient availability to crops (Khan et al. 2007), enhance the proliferation of beneficial bacteria (Priyadi et al. 2005, Javaid 2010), and prevent infection by pathogens (Compant et al. 2005). However, the additional microorganisms from commercial products have to be incubated and propagated in the lab and further applied. Their survival rate and their influences on plant and soil have been viewed skeptically (Watanabe et al. 2000, Chen et al. 2015).

In contrast, indigenous microorganisms may be optimally suited for survival and multiplication in significant amounts (Paluch et al. 2013). However, there is less information available on the ways to activate naturally occurring microorganism populations in soils. The use of CE produced by using plant extraction technology may be one method to activate indigenous soil microbial populations. CE derived from endophytes extracted from different plant species such as soybean (*Glycine max (L.) Merr.*), lucerne (*Medicago sativa L.*), the brown algae (ascophyllum nodosum) (Bestfarming systems Co.), and has been widely used recently. A new bio-molecular method for determining the presence of *nifH*, the gene harboured in entophytic bacterium for nitrogenase reductase from plant species has been developed (www.amazingcarbon.com) (Gao et al. 2015, Le et al. 2015). This study demonstrates that if CE activate indigenous microorganisms in the soil, particularly N-fixing bacteria or archaea that are not able to be cultured in the laboratory. Besides, increasing evidence suggests that isoflavonoids and flavonoids exuded from the root of many leguminous plants can activate rhizobium genes, which helps in the nodulation process (Peters et al. 1986, Brunetti et al. 2013). Combing microbial remediation with legume plant remediation is expected to be an economically and environmentally appropriate approach for soil amendment.

This study was undertaken to determine: (1) the significant effects of adding CE on the plant and microbial biomass, activity of the microorganism community, plant and soil nutrient; (2) combining CE and OM achieve the highest impacts on soil nutrient and lucerne growth; (3) CE stimulate the nodulation of legume as effectively as inoculating rhizobium bacteria. The results are expected to reveal the interactive effects of CE and OM application on plant species growth and soil properties, and aid in the selection of optimal fertilization approaches for soil quality

amendment. In addition, the experiments will enable explore the stimulative effect of CE on activating and enriching the abundance of indigenous microorganisms from the plant rhizosphere.

Materials and methods

Experimental design

A greenhouse experiment was conducted in the lab of prataculture science, Nanjing Agriculture University, Jiangsu province, China, from June 2015 to September 2015 as preliminary trial, and the visual evaluation was observed. To determine the actual effects of the treatments, the experiment was completely repeated from October 2015 to January 2016, and the second time data was recorded. The experiment was set up with 4 soil amendment treatments and 10 replications. The 4 treatments included: organic manure (OM), compost extract (CE), organic manure + compost extract (CEOM) and the control.

We collected the seed of lucerne from barren land in Xilinhot (43°38’ N, 116°42’ E), Inner Mongolia, China. The seed were sterilized firstly and germinated on 1.5% water-agar plates and transplanted into pots (height 15 cm, diameter 15 cm). Each pot filled with 1 kg of soil (dry weight equivalent) and 200 g double-washed quartz sand. Soil without containing any heavy metal was collected from the same study area and the soil had 1-year history for planting legume. The soil was taken from 15 cm depths with 5 cm diameter soil cores and evenly mixed, then applied to each pot. The characteristics of the soil were listed in table 1. Prior to seedling transplantation, OM was collected and mixed into the top layer of the potting applied at a rate of 5 g per pot. It was made of pig dung compost and amino acid fertilizer, the pig dung compost contained 30.1% organic matter, 3.0% N, 2.7% P_2O_5, 0.9% K_2O; And the amino acid fertilizer

86 contained 40.2% organic matter, 11.1% of amino acids, 3.4% N, 1.7%P_2O_5, 1.1% K_2O. Four of
87 ten strongest seedlings were finally left per pot. The CE consisted of alfalfa meal 15.9%
88 (weight/weight), barley grain 10.2%, barley straw 6.4%, wheat straw 4.3%, liquidized Fish
89 8.7%, kelp 39.5%, sulphur 0.3%, calcium carbonate 10.2%, and molasses 4.5%. The mixture
90 of CE was fermentated at < 50°C for 14 days in the incubator, then extracted, filtered, freezed,
91 dried. The final compositions (minimum guaranteed analysis) contained 16.9% of amino acids,
92 0.5% soluble potash (weight/weight), 0.06% calcium, 1.5% sulphur, 1.2% nitrogen,
93 0.3%phosphorus. 20 ml of CE (Best farming Systems Co.) (2 ml/pot for each application)
94 diluted with 5 L of water was sprayed onto the soil (the total application rate in the pot
95 experiment were comparable to agricultural practice with 750 ml /hectare). Four weeks later,
96 the CE was applied for second time (practical dosage at the manufacturers' recommendation)
97 and no more was applied after that.

98 A corresponding supplementary experiment on nodulation effects was conducted when the
99 seedlings were transplanted into additional pots (the same size as above). The seedlings in the
100 pots were inoculated with 6 ml liquid per pot with rhizobium strains (Rhizobium meliloti
101 dangeard- CX 107) cultured from nodules of the legume lucerne (provided by the Key
102 Laboratory of Agro-Microbial Resource and Application, Ministry of Agriculture, China
103 Agriculture University). Bacterial suspensions were diluted to have an optical density of 0.70
104 (λ= 600 nm), which was equivalent to a concentration of 3.10^8 bacteria/ml, as measured by the
105 Bradford method (Bradford 1976) on a yeast-mannitol culture medium (Vincent and Humphrey
106 1970).

107 Starting from 10. June 2015, the seedlings were grown under glasshouse conditions at 50-

70% relative humidity and a temperature regime of 20-25℃ during the day and night. Pots were watered to maintain soil water content between 20% and 25%. Eighty days later, seedlings of both experiments were destructively harvested, and separated into leaves, shoots and roots, measured their biomass, and examined for nodulation by counting and weighing the root nodules (roots were washed to remove the soil before checking).

Chemical analyses

Plant samples

Samples of the stem and leaves were milled to <0.2 mm and analyzed for total C and N. Plant total P content was determined using a UV/visible spectrophotometer (Beckman Coulter DU 800, USA). Plant total K content was measured using H_2SO_4-salicylic acid -H_2O_2-Se digestion (Zheng et al. 2012).

Samples of plant were ground to pass a 1 mm sieve and scanned twice using the Near-Infrared-Spectroscopy (NIRS) technique for forage nutritive value determination (Ren et al. 2016). Nutritional value was tested by evaluating crude protein (CP; nitrogen concentration×6.25), neutral detergent fibre (NDF), cellulose digestible organic matter (CDOM) and metabolisable energy (ME) (MJ/kg DM) (Schonbach et al. 2009). NDF was analyzed sequentially using the method described by (Vansoest et al. 1991), which used semiautomatic ANKOM 220 technology and was expressed with residual ash. CDOM value as a percentage of organic matter and ME value were calculated using crude ash (CA) and non-soluble enzymatic substance (EULOS). Detailed calculations are given in equations 1 and 2 below (Schonbach et al. 2009):

$$CDOM = 100\ (940 - CA - 0.62\ EULOS - 0.000221\ EULOS^2) / (1000 - CA); \quad (1)$$

130 $ME = 13.98 - 0.0147\ CA - 0.0102\ EULOS - 0.00000254\ EULOS^2 + 0.00234\ CP;$ (2)

131 Soil samples

132 Soil samples were separated in two portions, one portion was weighed to determine soil
133 bulk density by using foil sampler (volume =100cm^3) and dried at 105℃ for 24h to calculate
134 water holding capacity (%), another portion was stored at -22℃ for microbial abundance
135 analysis by real-time PCR (Gupta et al. 2017). Subsamples were sieved through 0.15mm and
136 analyzed for soil organic matter (OM), alkaline hydrolysable nitrogen (N), available
137 phosphorus (P), available potassium (K), and total C, N, P and K content. Soil OM content was
138 determined through the potassium dichromate external heating method (Blakemore et al. 1972).
139 The alkaline-hydrolysable diffusion method was applied to determine alkaline- hydrolysable N
140 content (Bremner et al. 1996). Available P was measured using the Olsen method (Blakemore
141 et al. 1972). Available K was measured with the flame photometry method (Blakemore et al.
142 1972). Total C was analyzed using the Kurmies determination (Blakemore et al. 1972). Soil
143 total N was analyzed using a Kjeltec analyzer (Kjeltec Analyzer Unit 2300, FOSS, Hillerød,
144 Sweden). Soil total P and K was measured the same way as plant total P. Soil pH was measured
145 in a 1:2.5 (soil: water) suspension.

146 Microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) as well as
147 microbial respiration (MR) were measured to compare differences in microbial colonization
148 and activity. MBC and MBN were estimated using the chloroform fumigation extraction
149 method (Frostegard et al. 2011). MR was measured in pre-incubated (24h at 28℃) samples by
150 determining CO_2 evolution over a period of 72 h (FAL, 1996). The abundance of viable bacteria
151 and fungi were analyzed according to the real-time quantitative PCR-DGGE (denaturing

152 gradient gel electrophoresis) of 16S rDNA method, which was performed using the ABI
153 7300real-time PCR system (Applied Biosystems, Foster, California, USA) with fluorescence
154 TaqMan@ probe detection (Le et al. 2015, Gupta et al. 2017).

155 **Statistical analysis**

156 For the complete randomized experiment design, one-way ANOVA (analysis of variance)
157 was used to determine differences among treatment for lucerne species growth or soil properties.
158 The LSD-test procedure was used for testing mean differences among control, OM and CE
159 treatments. The level of significance was $P < 0.05$. All the variables were statistically tested for
160 homogeneity of variance. All statistical analyses were carried out using SAS, Version 9.2 (SAS
161 Institute Inc., Cary, NC, USA).

162 **Results**

163 3.1. Biomass and nutritional value of lucerne

164 The shoot, root and total biomass of inoculated lucerne significantly increased ($P < 0.05$)
165 under CE application for 73.3%, 38.3% and 63.8%, respectively (Fig. 1). Compared with the
166 control, the three biomass of lucerne did not change under CE + OM treatment, but significantly
167 decreased from 17.5% to 26.6 % when OM was added. For non-inoculated lucerne, all three
168 biomass showed highest value with CE adding alone by increased around 46.0% to 85.3%, then
169 were second with CE + OM treatment. OM adding did not affect the biomass of non-inoculated
170 lucerne. The interactive effect between soil amendment treatments and plant nutritional values
171 were further analyzed using the ANOVA test (Table S1). CE and OM (Table 2) significantly
172 affected the nutritional values. The CP concentration of lucerne was significantly higher for the
173 OM treatment and CE treatment than the control. CDOM and ME showed the same trend in

response to treatments. The effect of CE and OM on NDF of lucerne significantly decreased from 66.18 to 52.45 g kg^{-1} DM and CE alone decrease NDF for 36.3%.

3.2. N, P, K contents in stems, leaves and soil

The N content of stems, leaves and soil decreased significantly in the order of CE + OM > OM > CE > control (Table 3, Table S1). The N contents of stem, leaves and soil in the CE + OM treatment were 40.4%, 35.0% and 91.4% higher than in the control treatment, respectively. The P content of stems was significantly higher in the three soil amendment treatments than in the control treatment. The P content of leaves and soils increased in the order: CE + OM > CE > OM > control. The K content of stems and soil was significantly higher in the CE + OM treatment than in the other treatments, and both the CE and OM treatments had a higher K content in stems than the control treatment. The soil amendment treatments significantly enhanced K content in leaves than in the control treatment. The K content of soil was decreased significantly in order of CE + OM > CE > OM > control treatment.

3.3. Biological and abiotic characteristics of soils

MBC and MBN showed a significant increase with treatments (Table 4, Table S1). In CE + OM treatment, MBC and MBN were 37% and 30% significantly higher than in the control, respectively. MBC was not significantly different between CE and OM treatments. Microbial respiration rates and metabolic quotients significantly increased in the order of CE + OM > OM = CE > control, at highest microbial respiration rate by about 82% and metabolic quotient by 28%. The numbers of soil bacteria and fungi showed an increase in treatments with CE, OM and CE + OM application, but were significantly higher with CE alone than OM. The application of CE and OM together strongly increased the amounts of bacteria in the

196 rhizosphere soil of lucerne.

197 Of the three soil amendments, only CE + OM significantly increased soil PH and water
198 holding capacity (Table 5, Table S1). The three soil amendment treatments significantly reduced
199 bulk density and increased organic matter, soil alkaline-hydrolysable N, available P content and
200 available K. Soil bulk density decreased significantly only between CE + OM treatment and
201 control.

202 3.4. Comparison between rhizobium nodulation among soil amendment treatments

203 The highest number of nodules and total nodule weight occurred in inoculated lucerne with
204 CE treatment (Fig. 2, Table S1 and S2). The number of nodules and total nodule weight of the
205 inoculated lucerne in the control treatment showed no significant difference to the non-
206 inoculated lucerne with CE application.With OM application, nodulation of lucerne decreased
207 significantly. Table S3 showed the effects of treatments on plant N and soil N of inoculated
208 lucerne.

209 **Discussion**

210 The shoot, root and total biomass and nutritional value of inoculated and non-inoculated
211 lucerne were significantly increased due to the application of CE alone and CE with OM
212 together. It indicates that CE acts better than OM for improving legume plant growth, because
213 nitrogen-fixation plant can be suppressed by external nitrogen adding. However, it does not
214 imply that OM can substitute for CE, because highly synergistic effects on plant nutrient and
215 soil properties were observed. Both soil biotic (MBC, MBN, MR) and abiotic (soil water
216 holding capacity, organic matter, alkaline N, available P and K) factors were improved, and thus
217 the plant growth was enhanced.

When CE was applied, 163.8% higher plant biomass occurred in the present study, which could be mainly attributed to the stimulation of the beneficial microorganisms (increased in numbers of bacteria and fungi, Table 4) by accelerating the decomposition of organic materials and increasing the release of nutrients (Javaid and Bajwa 2011). In contrast, the OM provided additional microorganisms and external nutrient into the soil (Higa 2001). These inoculated microorganisms have to compete with naturally occurring bacteria (Sherr et al. 1992). The interactions of different microorganisms may complement each other and further influence plant growth due to microbial diversity (van der Heijden et al. 2008). By stimulating the proliferation of beneficial bacteria and restraining harmful microorganisms through CE addition, some essential substances such as nucleic acids, amino acids and bioactive substances (e.g. hormones and enzymes) are synthesized. They can accelerate decomposition of lignin materials in the soil and mineralization of organic material as well as control soil diseases, resulting in the change in soil microbial parameters and physical and chemical characteristics as seen in this study (Kim et al. 2004, Javaid and Bajwa 2011). Besides, lucerne has been reported to produce the root-derived antimicrobials indole, terpenoid, benzoxazinone, flavonoid and isoflavonoid, which used for inhibiting soil-borne pathogenic bacteria (Dixon 2001) and thus enhance the symbiosis between plant and beneficial microorganisms. In soil organic amendment systems, the beneficial microbes activated by CE and added by OM accelerate the mineralization of the soil organic matter (Javaid 2011), which releases more nutrients for plant uptake (Flores et al. 1999).

Higher uptake of N, P and K by plant and higher soil content of N, P and K (Table 2) indicating that soil amendment treatments increased the availability of nutrients in the soil. In

240 this study, the soil had a low PH value indicating acidic conditions, which resulted in low
241 nutrient availability and low inorganic fertilization efficiency (Chen et al. 2015). The observed
242 increase in soil PH with applying CE and OM together demonstrated that combination of soil
243 conditioner and OM could amend the acidic soil by accelerating the rate of microbial processes
244 and thus releasing the nutrients in the acid soil quickly, as shown by other studies (Hati et al.
245 2006, Lee et al. 2009). Separately, they had no significant effects. Although both OM and CE
246 contain nutrient content, it is not necessary to be the major reason for nutrient increasing of
247 plant and soil. Because CE includes very less nutrient element but has greater effects on soil
248 and plant nutrient in comparing with OM. If the contribution of OM to soil and plant attribute
249 to additional nutrient adding, then CE need to have access to active soil nutrient availability by
250 using very less nutrient content.

251 The identical numbers of nodules as well as nodule weight of inoculated lucerne as non-
252 inoculated lucerne added with CE indicates the abilities of CE for activating indigenous
253 rhizobia (Le et al. 2015). The mechanisms for the maintenance of root-soil contact have been
254 attributed to root exudation (Le et al. 2015). The plant-soil feedback driven by root exudates
255 could initiate and manipulate signal traffic between roots and soil organisms (Keymer and
256 Lankau 2017). CE may stimulate root exudates, which could initiate one-way signals contacting
257 the chemical and physical soil properties to the roots, and regulate the symbiotic and protective
258 interactions with microbes (Jones et al. 2003, Johnson et al. 2015). They accumulate mainly
259 inducible antimicrobial compounds gathered in the roots (Flores et al. 1999, Dixon 2001). Many
260 indigenous microorganisms in the soil are not able to interact synergistically with plant species;
261 therefore, a number of antimicrobial root exudates are secreted. Le et al. (2015) confirmed that

some endophytic actinobacteria had the potential to enhance the growth of lucerne and its interactions inrhizobial symbiosis. The mechanism of how CE produces a stimulative effect on properties soil and plant growth need to be further explored.

Conclusions

This study demonstrated that CE application alone and in combination with OM enhanced lucerne biomass and nutritional value by improving nodulation and soil biotic and abiotic factors. The similar effects of CE and OM on plants and soils does not necessary mean that they can be treated as substitutes for one another. Rather, their combination provides a mode of soil amendment to improve soil quality effectively. CE synthesizes some kinds of active substrates that stimulate the activity of indigenous rhizobia in the soil rather than adding extra microorganisms into the soil as OM. Root exudation may help in regulating soil properties and root-microbe-soil interactions. However, the nature of their reactions in the soil need to be further explored.

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P Stoichiometry and Linkages to Ecosystem Functioning in the Inner Mongolia Grassland. PLoS ONE **7**:e51750.

Table 1 Soil properties in the study area

Soil property	pH	Bulk density (g/cm^3)	WHC (%)	OM (g/kg)	organic C (%)	total N (%)	total P (%)	C: N ratio (%)
value	5.88	1.61	25.73	13.25	12.12	0.83	0.16	14.60

The abbreviations are: OM (organic matter), WHC (water holding capacity).

Table 2 Effect of compost extract (CE), organic manure (OM) and CE + OM (CEOM) application on the shoot nutrient value of lucerne

Treatment	CP (g DM/kg)	NDF (g DM/kg)	CDOM (g DM/kg)	ME (MJ DM/kg)
Control	17.7c	66.18a	68.61b	9.84b
CE	23.3a	58.24c	77.77a	10.87ab
OM	21.4b	61.48ab	72.42b	10.09b
CEOM	24.6a	52.45bc	79.99a	11.01a

The abbreviations are: DM - dry matter; CP - crude protein; NDF - neutral detergent fibre; CDOM - cellulose digestible organic matter; ME - metabolisable energy; and MJ – joule per mole

Different letters within a column indicate significant differences ($P < 0.05$) between treatments. LSD multiple comparison was used.

Table 3 Effect of compost extract (CE), organic manure (OM) and CE + OM (CEOM) application on N, P, K content of lucerne stems, leaves and soil

Treatment	Stem (g/kg)			Leaf (g/kg)			Soil (g/kg)		
	N	P	K	N	P	K	N	P	K
Control	2.30c	0.72b	11.90c	3.77c	1.30c	7.51b	0.81c	0.18c	1.31c
CE	2.73b	1.17a	24.66b	4.89b	2.13b	13.39a	1.29b	0.26b	2.46b
OM	2.98b	1.17a	24.28b	4.19b	2.02b	13.36a	1.20b	0.24b	2.00b
CEOM	3.23a	1.22a	30.35a	5.09a	2.37a	13.17a	1.55a	0.35a	3.54a

Different letters within a column indicate significant differences ($P < 0.05$) between treatments. LSD multiple comparison was used.

Table 4 Effect of compost extract (CE), organic manure (OM) and CE + OM (CEOM) application on MBC, MBN, MR, QCO_2 and numbers of bacteria and fungi in rhizosphere soil of lucerne

Treatment	MBC (mg/kg)	MBN (mg/kg)	MR (mg/kg/d)	QCO_2 (mg C/kg)	Bacteria ($\times 10^6$copies/ml)	Fungi ($\times 10^6$copies/ml)
Control	303.21c	63.86c	49.01c	13.49c	22.5d	3.20c
CE	384.44b	70.60b	57.33b	14.92b	34.7b	4.33a
OM	340.73b	62.80c	59.81b	14.36bc	30.0c	4.01b
CEOM	414.50a	83.03a	89.00a	17.25a	38.5a	4.35a

The abbreviations are: MBC (microbial biomass Carbon), MBN (microbial biomass N), MR (microbial respiration rate), QCO_2 (metabolic quotient), N= 20.

Different letters within a column indicate significant differences ($P < 0.05$) between

treatments. LSD multiple comparison was used.

Table 5 Effect of compost extract (CE), organic manure (OM) and CE + OM (CEOM) application on soil characteristics

Treatment	pH	Bulk density (g/cm^3)	WHC (%)	OM (g/kg)	Alkaline N (mg/kg)	Available P (mg/kg)	Available K (mg/kg)
Control	5.90b	1.63a	25.38bc	14.22b	67.60c	4.02c	79.88c
CE	6.05ab	1.47b	28.48b	23.03a	102.46b	35.85b	159.79b
OM	5.95b	1.46b	25.87c	22.53a	110.63b	50.08a	204.70a
CEOM	6.33a	1.32b	35.17a	25.37a	128.20a	52.83a	214.37a

The abbreviations are: OM (organic matter), WHC (water holding capacity)

Different letters within a column indicate significant differences ($P < 0.05$) between treatments. LSD multiple comparison was used.

Fig. 1 Shoot biomass, root biomass and total biomass of inoculated (a) and non-inoculated (b) lucerne under different soil amendment treatments: control, compost extract (CE), organic manure (OM) and compost extract + organic manure (CEOM).Bars represent the standard errors, n= 20

Fig. 2 Effect of soil amendment treatments: control, compost extract (CE), organic manure (OM) and compost extract + organic manure (CEOM) on inoculated and non-inoculated nodulation of lucerne plants. Left: number of nodules; Right: total nodule weight. Straight line showed the same level between inoculated control and non-inoculated CE adding treatment. Bars represent standard errors, n = 20

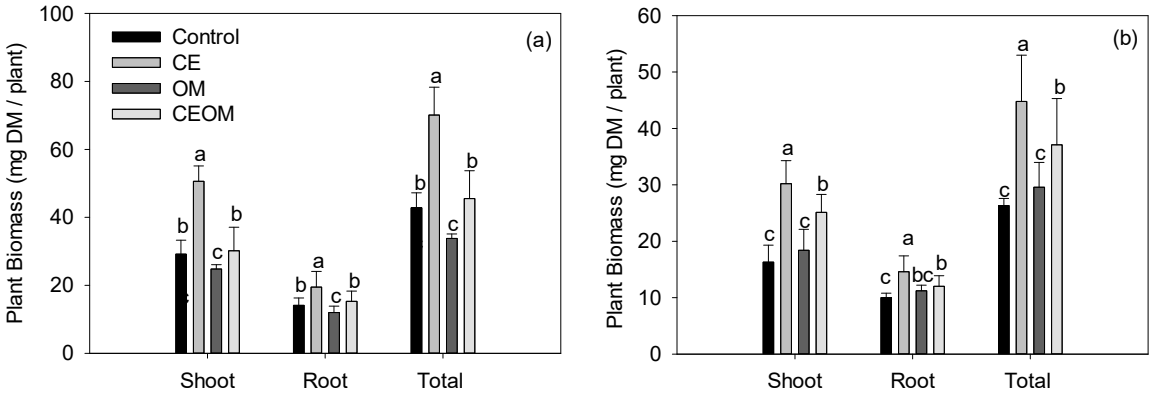

Fig. 1

Fig. 2