Effect of different machinery and rolling times on the microbial activity of reclamation soil in coal area

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Abstract Mechanical construction will put influence on the biological characters of reclaimed soils, as well as the soil quality. In order to explore the changing rule of soil microbial quantity and respiratory capacity under different construction machineries and rolling times, and find the optimal processing conditions, an experiment was set up and a simulation experimental area was chosen, in which we simulated the main types of reclamation in coal mine area. After 2 years' natural aging, we collected surface soil samples (0–20 cm) that can be used for experimental analysis. The result shows that changing rules of soil biological factors are different with different construction machineries, and soil properties are closest to the normal soil when adopting the combination of "crawler dozer \times 5 compaction times" and "dump truck \times 3 compaction times", which shows that the soil quality is better under this condition.

Keywords Construction machinery · Compaction times · Reclamation soil · Microbial quantity · Respiratory capacity

1 Introduction

Due to the coal mining, large land areas are damaged in China every year. And so far, the number of various industry and mining waste land has exceeded 4 million hectares annually (Wang et al. 2009a). Our national reclamation technologies of coal mining subsidence area are various, but the level of reclamation quality depends on the soil physical structure, chemical property and biological fertility, and the biological fertility is measured by soil microorganism quantity and activity in general (Fan et al. 2011).

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In recent years, with the development of modern molecular biology and analytical chemistry as well as the advancement of information technology, the study of soil microorganisms has become a new hotspot of soil science. Being the living organisms, soil microorganisms decompose the soil organic matter and participate in all kinds of active enzyme metabolic processes in the soil. They are sensitive to changes of environment by playing an irreplaceable role in maintaining soil ecological structure, keeping the balance of soil nutrients and protecting moisture and fertilizer (Bian 2005; Wang et al. 2006; Lin et al. 2010). Since soil respiration indicates life activities in the soil, it is the main link of terrestrial ecosystem carbon cycle (Wang et al. 2007).

Using different construction machineries to compact different times, and enabling each plot has different degrees of compaction, are two ways that have different impacts on soil bulk density, water permeability, air permeability, porosity and other physical properties. Those factors will affect the activity of microbial community and the growing conditions of vegetation growth, as well as the quantity and activity of microorganisms ultimately. Many scholars at home and abroad study and analyze microbial

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activity from different aspects: such as Tang et al. who studied the influence of microorganism quantity and soil respiration under different land utilization types in Dongting Lake region, Zhang et al. who made researches on the relationship among the soil nutrient, pH and microbial quantity, Hamza and Anderson who studied soil nitrogen, phosphorus, potassium, organic matter and microbial activity from the perspective of soil compaction (Hamza and Anderson 2005; Schaffer and Attinger 2007; Zhang et al. 2011; Tang et al. 2013). However, analyzing the activity of microorganisms from the perspective of construction machinery and construction times is not common in China. This research built an experimental area in the field environment, studied the soil microorganism activity under setting conditions, and discussed the change of microorganism quantity under different conditions, and then found the optimal condition for microorganism growth.

2 Materials and methods

2.1 Natural conditions of experimental area

The experimental area is located in the south campus of Shandong Agricultural University experimental field, whose geographical position is 36.20° north latitude, 117.08° east longitude. This area belongs to warm semihumid continental 'monsoon climate region, with an annual average temperature 13.2 °C and annual average rainfall 803.7 mm.

2.2 Experiment design

Based on the Natural Science Foundation of China in July 2011, this experiment was set up and the simulation experimental area was chosen, and simulating main types of reclamation in coal mine areas was selected. The construction waste, fly ash and coal gangue being filler material, NT855-B280 crawler dozer and Dongfeng EQ 3060 dump trucks being the reclamation machinery, the simulation compaction process was carried out for five

times (one, three, five, seven, nine times). With natural setting soil as the contrast, there are 31 pieces of sample areas totally. The experimental design is shown in Fig. 1.

Every piece of sample area is a one metre cube with 40 cm filler at the bottom and 60 cm silty loam in the upper. The experiment construction is shown in Fig. 2. After 2 years' natural aging, we collected surface soil samples (0–20 cm) of experimental area on July 18, 2013, which are used for experimental analysis. In order to avoid the marginal effect, the soils we collected are at the range of "0.5 m \times 0.5 m" in the middle of the sample area.

2.3 Sample processing

For the soil microbiota analysis, the dilute flat surface smear method is adopted. The study analyzed the bacteria with beef extract peptone medium, fungi with Martin's rosebengal medium, and actinomycetes with improved Gause's No.1 medium (Nanjing Institute of Soil Microorganisms 1985), and each group of samples conducted experiments three times repeatedly, then abandoned the data whose reproducibility were bad, and calculated the average as the soil microorganism quantity of this group of samples. The determination of soil respiration adopted the static alkali absorption method, and the specific approach was to put a container filled with NaOH solution exposure after a period of time, when part of the lye absorbing CO₂ and forming carbonate, we can calculate the residual amount of lye by the neutralization titration method, thus getting the amount of CO₂ released from the soil within a certain time according to the corresponding formula (Yan et al. 2010). The data are processed by using Excel software first, and then correlation analysis was carried out by using the SPSS.

3 Data analysis and results

3.1 The change of the microbial quantity under different soil compactions

The condition of the soil microbial growth is affected by various environmental factors in the soil, and it is one of

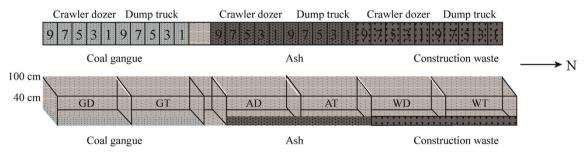


Fig. 1 The experiment design

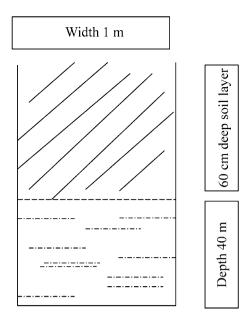


Fig. 2 The experiment construction

the most sensitive indicators for the change of the soil quality at the same time (Chen et al. 2007), whose number and distribution can reflect the subtle change of soil ecological system. To some extent, different construction machineries and rolling times can make different soil compaction degrees, and then cause the change of soil bulk density, porosity, water permeability, air permeability and other physical properties. All these factors above will affect the number of soil microorganisms, as shown in Table 1, they are the number of different soil microorganisms under different construction machineries and compaction times.

In general, we can see from Table 1, that the microbial quantity size order is bacteria > actinomycetes > fungi, no matter how many times construction machineries compact;

in addition, the soil microbial number of experimental plots and the total number of microorganism are all below the contrast level (except for crawler dozer, three times compaction), and this is because that compared with natural sinking soil, mechanical compaction can increase soil bulk density, which affects the water and air permeability, porosity and root penetrative resistance, thereby can impact the growth and development of root system. These cases will affect nutritional source of the soil microorganism additionally to some extent. On the other hand, for the aerobic saprophytic microorganism in the soil, the compacted soil cannot provide enough oxygen for them, and all of these factors affect the microbial activity indirectly (Wang et al. 2009b).

3.1.1 The influence of different construction machineries on soil microbial quantity

The main function of bacteria is to decompose organic matters (Yan 1998), and as is shown in Table 2, the bacteria plays a dominated role in the soil microbial community, accounting for about 70 % of the total. For the plot using dump truck to compact, the proportion of bacteria is 70.71 %, but for that using crawler dozer, the proportion is 71.71 %. Actinomyces can transform the soil organic matter and is also the important antibiotics producing strain at the same time, the quantity of actinomyces ranks secondly in the microbial community only behind the bacteria with a proportion of 29.24 % for the plot using dump truck to compact and 28.25 % for the plot using crawler dozer. The quantity of fungi is inferior to bacteria and actinomycetes in the soil microbial community: less than 1 %, and the proportion is 0.05 % for the plot using dump truck, and for the plot using crawler dozer to compact, its proportion is 0.04 %.

 Table 1 Microbial quantity of different construction machineries and rolling times

Construction machinery	Compaction times	Bacteria quantity (10 ⁶ cfu/g)	Fungi quantity (10 ⁶ cfu/g)	Actinomyces quantity (10 ⁶ cfu/g)	Microorganism quantity (10 ⁶ cfu/g)
Dump truck	1	7.08	3.92	3.21	10.29
	3	8.67	6.96	3.56	12.24
	5	8.58	6.57	3.74	12.33
	7	7.33	6	2.72	10.06
	9	6.33	3.74	2.48	8.81
Crawler dozer	1	7.73	4.09	2.02	9.75
	3	11.42	6.82	3.56	14.99
	5	8.22	6.97	4.91	13.14
	7	7.61	4.01	3.35	10.97
	9	7.58	2.33	2.93	10.51
СК	Natural sinking	9	8.27	4.63	13.6

Notes In addition to the CK, the rest data are the average from the plots regarding to construction waste, fly ash and coal gangue as filler

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Construction	Bacteria		Fungi		Actinomyces		
machinery	Quantity (10^6 cfu/g)	Proportion (%)	Quantity (10^3 cfu/g)	Proportion (%)	Quantity (10^6 cfu/g)	Proportion (%)	
Dump truck	7.598	70.71	5.438	0.05	3.142	29.24	
Crawler dozer	8.512	71.71	4.844	0.04	3.354	28.25	

 Table 2 Microbial quantity and proportion of different construction machineries

Notes The microbial numbers are averages of microbial quantity under different compaction times

In addition, the microbial quantities are different when using different construction machineries to compact; for the bacteria and actinomycetes, the quantitative size order is: crawler dozer > dump truck. When using two kinds of machineries to compact at the same time, the dump truck makes a bigger soil compaction degree, so the plots that using dump truck to compact have a bigger soil bulk density, worse soil permeability and stronger root penetrative resistance, and thus these cases hinder the growth of bacteria and actinomycetes, expressing the less quantities of the bacteria and actinomycetes. For the fungi, the quantitative size order is: dump truck > crawler dozer, which is because for the plots that compacted by dump truck, the growth of the bacteria and actinomycetes are hindered, so the fungi acquires more nutrition and bigger living space, and at this time, the effect of soil bulk density is not significant, thus the quantity of the fungi from the plot that compacted by dump truck is higher than that from the plot compacted by crawler dozer slightly.

3.1.2 The influence of different compaction times on soil microbial quantity

Using different machinery to compact for one, three, five, seven, nine times respectively, the quantity of the soil microorganism increases and then decreases, presenting a variation of inverted "V" type. When using dump truck to compact, the quantities of soil bacteria and fungi reach the peak at the three times compaction, are 8.67×10^6 and 6.96×10^3 cfu/g respectively, and the quantity of actinomyces reaches the peak at the five times compaction, is 3.74×10^6 cfu/g; when using crawler dozer to compact, the quantity of soil bacteria reaches the peak at the three times compaction, is 11.42×10^6 cfu/g, and the quantities of soil fungi and actinomyces reach the peak at the five times compaction, are 6.97×10^3 and 4.91×10^6 cfu/g. Total quantities of soil microorganism reach the peak when using dump truck to compact three times and using crawler dozer to compact five times, are 12.33×10^6 and 13.14×10^6 cfu/g respectively. In addition, total microbial quantities are closest to the contrast when using dump truck to compact three times and using crawler dozer to compact five times, indicating that microbial activities of those cases are most similar to the natural soil. The soil bulk density increases, the porosity decreases and the permeability reduces as the compaction continues, and the growth of the microorganism is limited because of a short source of nutrient and oxygen; when the compaction times are not enough, the soil texture is too loose and the pore ratio is irrational, then there are too many large pores, which go against the rooting of soil plants, thus impacting the growth of root microorganisms (Fig. 3).

3.2 The influence of different soil compaction on soil respiration intensity

Soil respiration refers to the carbon flowing from soil to atmosphere in the form of CO_2 , including three biological processes (plant root respiration, soil microbial respiration, soil animal respiration) and a biology process (chemical oxidation process of carbon material). But it is generally believed that the soil respiration is mainly derived from an autotrophic respiration of plant roots and a heterotrophic respiration of soil microorganisms. Besides, the autotrophic respiration of plant roots includes living root tissue respiration, symbiotic root microorganism respiration, and the CO_2 produced from root exudates as well as the decomposition of dead roots (Yan et al. 2010). To a certain extent, the soil respiration intensity can measure the strength of the microbial activity.

According to the soil respiratory capacities under different compaction machineries, the averages of soil respiratory capacities from the plot that compacted by dump truck and crawler dozer are 30.478 mg CO₂/100 g soil and 52.232 mg CO₂/100 g soil respectively, and the comparison of soil respiration intensity is crawler dozer > dump truck, which is caused by physical and biological factors. On one hand, the compaction degrees of the plot that compacted by dump truck are bigger, which makes the soil permeability worse, and goes against the exhaust of CO₂; on the other hand, the bigger compaction degrees cause the larger soil bulk density, which hinders the rooting of plants and the growth of microorganisms, makes the autotrophic respiration of plant roots and the heterotrophic respiration microorganisms weak, and results in less soil respiration.

In addition, we can see from Table 3, the soil respiration intensities from plots compacted by machineries are inferior to natural soil except some special cases, indicating

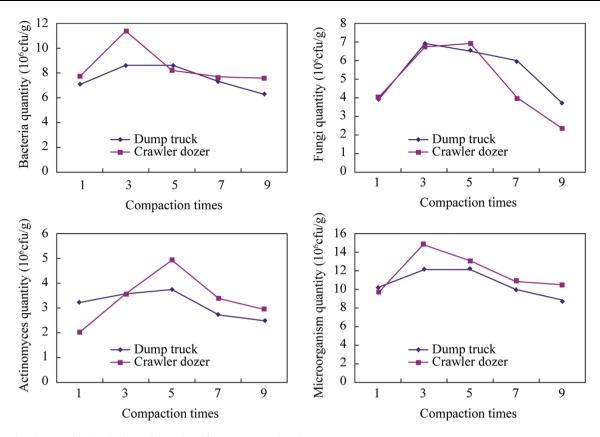


Fig. 3 The change of microbial quantity under different compaction times

 Table 3 Soil respiratory capacity under different construction machineries

Construction machinery	Compaction times	Soil respiratory capacity (mg CO ₂ /100 g soil)	Average (mg CO ₂ / 100 g soil)
Dump truck	1	20.56	30.478
	3	34.32	
	5	53.17	
	7	26.01	
	9	18.33	
Crawler dozer	1	22.56	52.232
	3	66.05	
	5	68.31	
	7	56.38	
	9	47.86	
CK Natural sinking		55.31	55.31

that the soil compactions caused by mechanical compaction have a bad effect on soil respiration, so the compaction times should be paid attention to when using machinery.

As is shown in Fig. 4, when using different machineries, the soil respiratory capacity increases and then decreases with the increasing of compaction times, presenting a variation of inverted "V" type. When using dump truck

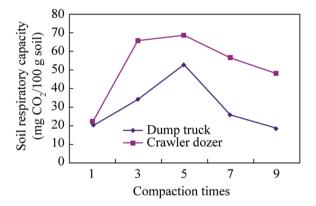


Fig. 4 The change of soil respiratory capacity under different compaction times

and crawler dozer to compact, at five times the soil respiratory capacities reach the peak, which are 53.17 mg $CO_2/$ 100 g soil and 68.31 mg $CO_2/100$ g soil. When the soil respiratory capacity reaches the peak and continue the compact, the soil bulk density will increase and affect the growth of plants and microorganisms, and at the same time the bad permeability will also hinder the exhaust of CO_2 , and reduce the soil respiratory capacity; before reaching the peak, the soil texture is too loose on the contrary, the

Microbial quantity and respiratory capacity	Bacteria quantity	Fungi quantity	Actinomyces quantity	Microorganism quantity	Soil respiratory capacity
Bacteria quantity	1	0.618	0.409	0.920**	0.658*
Fungi quantity	0.618	1	0.623	0.728*	0.42
Actinomyces quantity	0.409	0.623	1	0.735*	0.763*
Microorganism quantity	0.920**	0.728*	0.735*	1	0.818**
Soil respiratory capacity	0.658*	0.42	0.763*	0.818**	1

Table 4 The correlation between microbial quantity and soil respiratory capacity

* Indicates significant correlation

** Indicates extremely significant correlation

pore ratio is irrational and the big pore is over much, making soil plant roots feeble and affecting the normal growth of microorganisms, so the soil respiratory capacity is low.

3.3 The correlation between soil microbial quantity and respiratory capacity

The Table 4 shows that the correlation between microbial quantity and respiratory capacity is significantly positive (P < 0.01), with a correlation coefficient 0.818; the correlations between bacteria quantity and respiratory capacity as well as actinomycetes quantity and respiratory capacity are all positive (P < 0.05), correlation coefficients being 0.658 and 0.763 respectively; the correlation between fungi quantity and respiratory capacity is smaller, with a correlation coefficient 0.42.

4 Conclusions

The research shows that:

- (1) The order of soil microbial community is: bacteria > actinomycetes > fungi, and the bacteria accounts for about 70 % of the microorganism, the actinomyces for about 30 % and the fungi for less than 0.05 %.
- (2) Quantities of bacteria and actinomycetes in plots compacted by crawler dozer are more than those compacted by dump truck, and the quantity of fungi is just the opposite. But overall, the total quantity of microorganism in the plots compacted by crawler dozer is more than those compacted by dump truck. With the increase of compaction times, the quantity of microorganisms increases firstly and then decreases just like an inverted "V". The quantity reaches the peak when the plot is compacted three times by dump truck or five times by crawler dozer, and at this time, the quantity of microorganisms is nearest to the natural soil.

- (3) It is similar to microorganisms that with the increase of compaction times, the change of the soil respiratory capacity presents an inverted "V". Its amount reaches the peak when compacting three times; and the soil respiratory capacity in the plot compacted by crawler dozer is larger than those compacted by dump truck.
- (4) A correlation between microbial quantity and respiratory capacity is significantly positive (P < 0.01), with a correlation coefficient of 0.818; the correlations between bacteria quantity and respiratory capacity as well as actinomycetes quantity and respiratory capacity are all positive (P < 0.05), with the correlation coefficients being 0.658 and 0.763 respectively; and the correlation between fungi quantity and respiratory capacity is smaller, with the correlation coefficient being 0.42.
- (5) Adopting the combination of "crawler dozers × compacting five times" and "dump trucks × compacting three times", the soil microbial activity is the strongest and nearest to normal.

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