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Exploring the Regulatory Role of Wood Vinegar on Heavy Metals (Copper, Zinc) Before and After Cow Dung Composting

Zhiyu Li, Ling Zhou, Fei Liu, Xuening Luo, Jinlong Sun, Cuilan Qin and Leiyuan Wang

ABSTRACT

In recent years, livestock manure pollution problems have become increasingly prominent phenomenon. It has brought enormous environmental pressure to our country and also provided the technical support for effective cow dung composting and high-fiber waste treatment. The experiment adopts the biomass wood vinegar as cow dung compost additives to study before and after the changes of heavy metal as Copper and Zinc. A total of six treatments, respectively, 0.20, 0.35, 0.50, 0.65, 0.80% of wood vinegar and control group. Marked them as T1, T2, T3, T4, T5 and CK. The passivation effect of Copper was T5> CK> T4> T3> T1> T2 and the best passivation effect of Copper was to add 0.80% wood vinegar, the value was 18.78%. The passivation effect of Zinc was T2> T5> T4>T1>T3> CK and the best passivation effect of Zinc was to add 0.35% wood vinegar which was 8.25%. The passivation effect of Copper and Zinc appeared on fluctuations before and after the composting process. Meanwhile, in order to further study, to prevent "Concentrated effect", "Chelation", "Dilutioneffect" and environmental change common giveriseto fluctuations further occurrence during the composting process. Ensure the stability of the passivation effects, minimizing interference, trying to increase the volume of compost, expanding treatment of wood vinegar gradient and extending the composting period. To prevent the secondary factor was greater than the passivation effect of wood vinegar treatment composting. It could achieve healthy and sustainable development of agriculture and environment.

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INTRODUCTION

With the development of economy and the improvement of people's living standard, livestock and poultry industry developed rapidly. But in livestock and poultry production, in order to prevent the occurrence of animal diseases, promote animal growth and add some heavy metals such as Copper and Zinc were more commonly used in the feed (Li et al., 2012). As the animal manure has not been processed and utilized effectively, a large number of livestock manure pollutants discharged into the environment which brought enormous environmental pressure to our country. The key to determine whether heavy metal is harmful or useful for environment, human being and animal is its biological effectiveness (Blais et al., 1992). The biological effectiveness of heavy metals is closely related to heavy metal forms and the soluble state of heavy metal is the main form in heavy metal mobility. It is the easiest way for plants absorbed and assimilated and also it is the greatest potential form of pollution in the food chain (Li and Zhang, 2001). At present, heavy metal pollution control methods are composting, passivation method, drain leaching and chemical method (Li et al., 2013). But the biomass wood vinegar (Zhou et al., 2009) as additive to process the heavy metals of livestock manure is rare. The use of the biomass wood vinegar for heavy metals passivation processing in cow dung has not been reported. In addition, our country is rich in agricultural biomass resources. There are about 700 million tons crop straws each year, the forestry residues (excluding fuel wood forest) approximately 27 million m³ which equal to 10 million tons standard coal (Li

each year, the forestry residues (excluding fuel-wood forest), approximately 57 million m³ which equal to 10 million tons standard coal (Liu *et al.*, 2006), cotton stalks of Xinjiang alone can reach annual production of 600~750 million tons (wet material) (Li *et al.*, 2007) according to the statistics. These residues provide sufficient raw materials for wood vinegar. The utilizing of wood vinegar regulates pollution problems in animal manure composting process, not only can achieve efficient use of agricultural and forestry residues resources but also solve the livestock manure pollution problems at the same time and have great benefits to ecology, resources and agricultural sustainable development.

MATERIALS AND METHODS

Experimental materials: Raw material was taken from the cattle farm, College of Animal Science, Tarim University which was pre-composting of manure. The sawdust was from the timber processing plant outside school. The wood vinegar additives was made by cotton stalks in the biomass pyrolysis experimental device at 550°C, filtered and stored 10 days. Cotton stalks were provided by Alar farms cotton growers (Table 1).

Composting program: The experiment was conducted in the breeding base, College of Animal Science, Tarim University. A total of six compost processing experiments (Table 2) and the dosage of wood vinegar was the ratio of wood vinegar and cow dung composting fresh weight. Once the amount of wood vinegar was confirmed, it was necessary to add 1500 mL of distilled water for dilution in addition. Meanwhile, 1500 mL of distilled water was added to the control group.

Composting technology: One of the key factors in compost operation is the C/N ratio of the composting material, this value is generally more appropriate for 20~30 (Li *et al.*, 2004). The cow dung and sawdust by uniformly mixing ratio of 1.5:1, so that the C/N value was 20.9, keeping the **moisture content** at about 61% (Wu *et al.*, 2003) and adjusting the **moisture content** of medium-term. The static forced ventilation+pile-turning method was used in this experiment. Ventilation once a day at 11:00 am, every ventilation last about 30 min, during the first period. At the 20th day, ventilation should be stopped and pile-turning should be executed. At the 26th day, pile-turning once again until the end of the composting, the composting was taken about 30 days in total.

Device composting: The device was made independently. It was a plastic bucket with 1.1 m effective diameter and the height was 0.37 m in general. At the bottom of the plastic bucket a hole with 2 cm diameter was made. A PVC pipe with 2 cm diameter was inserted into the hole and a 2 cm high support bracket was putted inside the plastic bucket. On the top of the support there was a grate with a layer of woven plastic membrane which has good air permeability. The PVC pipe was connected to a blower for the ventilation of oxygen. Figure 1 shows a small forced ventilation aerobic composting device.

Main instruments: PHS-3C pH meter, Analysis of electronic balance (FA1004), Drying oven (GZX-9140MB), Box-type resistance furnace (SX-2.5-10), Biomass pyrolysis experimental device (BRES06-1), SW-2 Microwave digestion system (SpeedWave 2), Atomic absorption spectrophotometer (AA800).

Sampling methods: Compost samples were got from before and after the process of composting. Each sub-sample was sampled randomly from the inner, middle and outer layer of the compost at 11:00 am. Then three sub-samples were mixed into a sample to be tested. Multi-level multi-point sampling was used and fully mixed, samples were samped before pile-turning. The samples were oven at 105°C, baked for 24 h and then grinding by 100-mesh sieve when the weight was dried to constant (Yang *et al.*, 2003), stored for testing under 4°C in the end.

Table 1: Basic physical and **chemical properties** of cow dung composting raw materials

Raw materials	Organic matter (%)	Total nitrogen (%)	C/N ratio	Specific conductance (mS cm ⁻¹)	Moisture content (%)	pH
Cow dung	42.20	3.53	20.62	2.89	45.00	7.29
Sawdust	47.51	0.28	292.50	0.84	6.31	6.00

Table 2: Composting experiment with different treatments

Treatment	Name	Abbreviations
Cow dung+0.00% wood vinegar	Control group	CK
Cow dung+0.20% wood vinegar	Treatment 1	T1
Cow dung+0.35% wood vinegar	Treatment 2	T2
Cow dung+0.50% wood vinegar	Treatment 3	T3
Cow dung+0.65% wood vinegar	Treatment 4	T4
Cow dung+0.80% wood vinegar	Treatment 5	T5

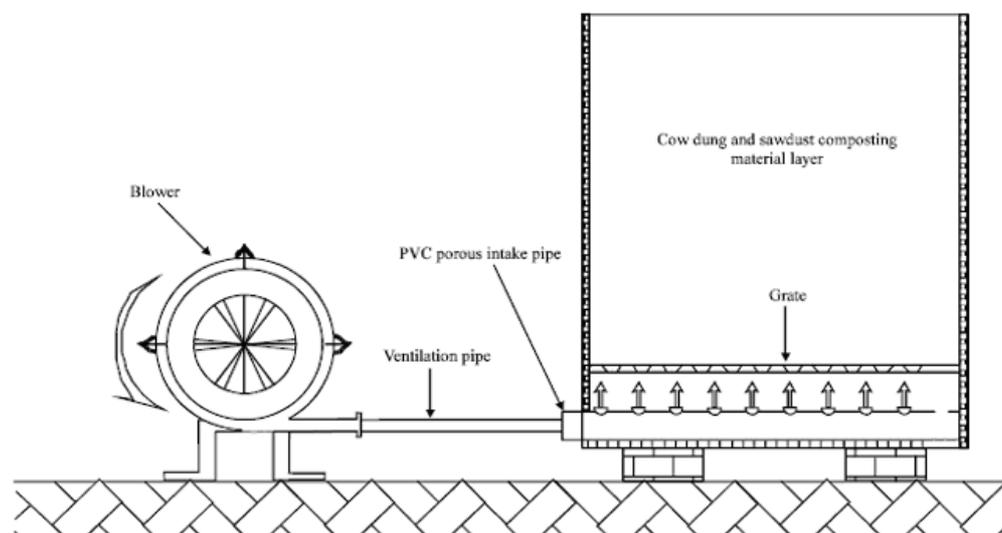


Fig. 1: Small-scale forced ventilation aerobic composting device (drawing by myself)

Determination methods: Organic matter was determined by burning method (CJ/T 96, 1999), Total nitrogen determined by the Kjeldahl method (NY 525, 2002), pH was determined by pH meter method, EC by Conductivity method (LY/T 1251, 1999) and Moisture content were determined by Vacuum oven method (GB/T 8576, 2010).

Total content of copper and zinc test solutions: The sample was dried to a constant weight at 60°C and cooled naturally. Then the samples should be stored in a polyethylene bottle after grinding through the 100-mesh sieve. Weighed 0.5 g (accurate to 0.0002 g) sample, adopted HNO₃: HCl: HF = 1:1:2 microwave (wet) digestion (Scancar et al., 2000). With deionized water constant volume after filtration.

DTPA extraction copper, zinc effective state test solutions: After digestion of samples under test and the extracting agent by solid-liquid ratio (w: v) 1: 5. The extracting agent was the added 0.005 mol L⁻¹ DTPA (Diethylene triamine pentacetate acid), 0.1 mol L⁻¹ TEA (Trolamine). After the solution was adjusted to pH 7.30 it needed the mechanical oscillation for 2 h (Lindsay and Norvell, 1978; Navarro et al., 1993). DTPA extractable copper, zinc partition coefficient calculation formula was (Zheng et al., 2005):

$$\text{Partition coefficient (\%)} = \frac{\text{Concentration of DTPA extractable heavy metals}}{\text{Total concentration of heavy metals}} \times 100$$

To measure the passivation effect of the composting process by changes of copper and zinc partition coefficient differences and this method has been adopted by many researchers (Zhang et al., 2006; Zhao et al., 2007). Partition coefficient differences were obtained by the initial partition coefficient subtracts the partition coefficient of arbitrary time sample in the composting process.

The determination of copper and zinc contents, atomic absorption spectroscopy method was used to measure the total content of heavy metal copper, zinc and the contents of DTPA extraction.

Data processing and analysis: Data processing and statistical analysis adopted SPSS13.0 statistics software. This experiment used a single factor random variables analysis of variance. Significance test was conducted at the level of $\alpha = 0.05$. The SPSS13.0 statistical software was used for data analysis of variance and multiple comparison on the determination results, MATLAB software was used for plotting graphs.

RESULTS

Heavy metal copper analysis: The DTPA extraction of heavy metals was well correlated with their **bioavailability**. DTPA was the most common reagents used for such form's extraction (Rauret, 1998). So this experiment studied the effects of wood vinegar to heavy metal water soluble before and after composting. Due to the evaporation of moisture, the degradation of material, **organic matter**, CO₂ and volatile material losses and the metabolism produced a lot of heat so that it induced heat and **mass transfer** in the composting process. Thus leading to the pile body becomes smaller and decreasing the quality of **dry matter** and creating a "Concentrated effect" (Zhao et al., 2007; Yang et al., 2006). Since the content of initial moisture and easily degradable **organic matter** were too much high at the early stages and led to the heavy metal "Concentrated effect" stronger, so the total concentration of heavy metals increased after composting. In conclusion, the concentration of extractable by DTPA before and after composting could not objectively reflect the practical issues. So the change of relative content (the ratio of distribution coefficient that DTPA-extractable concentrations and their total concentration) was adopted to have a better reflect on the passivation effects of wood vinegar to heavy metal effectiveness before and after composting.

As the results shown, there was a striking difference of heavy metal copper with wood vinegar treatment between treatment group and the control group according to the F-test results (Table 3). In addition, multiple comparison with Duncan method (SSR method) was used in order to further illustrate the relative roles size between the concentration of wood vinegar (Table 3). The analysis results were shown in Table 4.

These results demonstrate convincingly that using wood vinegar treatment the heavy metals copper got good results. When less than 0.20% wood vinegar added, the passivation effect of treatment group was lower than the control group. But, the passivation effect increased linearly with the content of wood vinegar. Since wood vinegar itself contains **organic acids**, aldehydes, ketones, alcohols, phenols and other organic compounds and their derivatives and heavy metals can be diluted and neutralized by wood vinegar. Wood vinegar could also promote the transformation of heavy metal forms which could transform heavy metals into the various salts which have lower compounds of biological effectiveness. This means that the larger additive proportion, the more heavy metal converted into compounds, lower **bioavailability** and the less danger.

Partition coefficient of copper after the composting process generally was lower than before composting in this experiment. Partition coefficient reduced respectively 8.15, 1.09, -0.12, 7.18, 7.36 and 18.18%. Compared with the control group when the added quality of wood vinegar was smaller, the partition coefficient was lower, conversely higher. Experimental results showed that the wood vinegar had good passivation effect for copper and the best passivation in treatment 5 was 18.18%. Partition coefficient appeared on the positive and negative situation which means the passivation effect of copper under treatment of the wood vinegar had small fluctuation (Table 3, Fig. 2 and 3).

Table 3: Changes of the total concentration of heavy metals copper and DTPA extractable in cow dung samples before and after the wood vinegar processing composting

Treatments	Total concentration (mg kg ⁻¹)	DTPA-copper concentration (mg kg ⁻¹)	Partition coefficient (%)	Partition coefficient difference (%)
CK				
Before composting	3.66±0.06	1.14±0.03	31.03	
After composting	4.71±0.06	1.08±0.03	22.88	8.15 ^a
T1				
Before composting	4.05±0.06	0.92±0.02	22.62	

After composting	3.02±0.06	1.09±0.02	21.22	1.07
T2				
Before composting	3.94±0.05	0.82±0.02	20.81	
After composting	5.21±0.12	1.09±0.02	20.93	-0.12 ^a
T3				
Before composting	3.90±0.11	0.82±0.02	21.11	
After composting	4.40±0.12	0.61±0.02	13.93	7.18 ^b
T4				
Before composting	4.21±0.06	0.89±0.02	21.24	
After composting	4.78±0.19	0.66±0.02	13.88	7.36 ^b
T5				
Before composting	3.96±0.05	1.24±0.02	31.31	
After composting	5.08±0.12	0.64±0.01	12.53	18.78 ^a

Data is mean of 3 replicates±standard deviation. The negative number of the partition coefficient difference indicated: The extractable content increased in the proportion of the total content after the composting, the positive number indicated a decrease. In the same column with the same letter indicated no significant differences between treatments at $p = 0.05$

Table 4: Significant analysis of different treatments

Treatments	Treatments	Treatments	Mean differences significance test
CK	T5	CK, T1, T2, T3 and T4	Significant
	T4	T3	Not significant
	T4, T3 and CK	T1 and T2	Significant
	T1	T2	Not significant

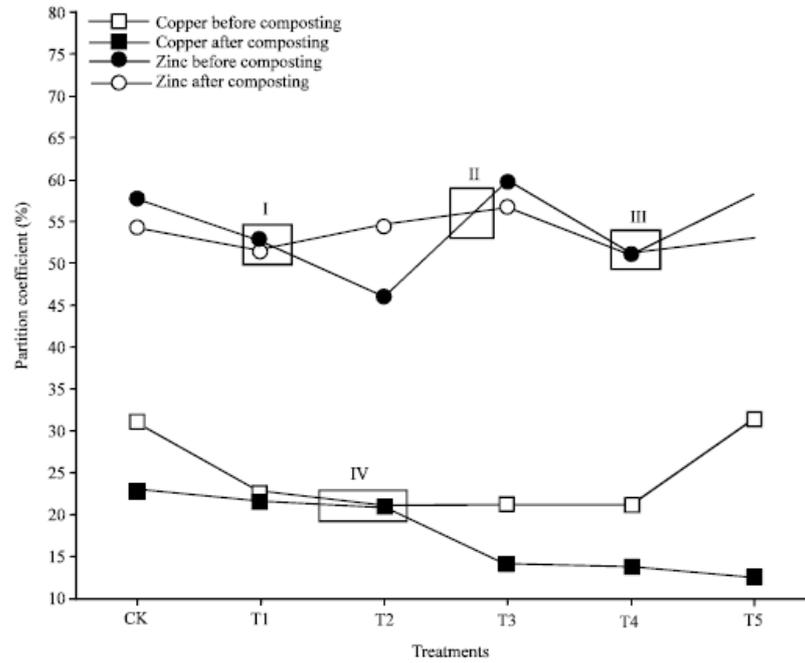


Fig. 2: Partition coefficient changes of heavy metal copper and zinc in cow dung samples before and after wood vinegar processing composting

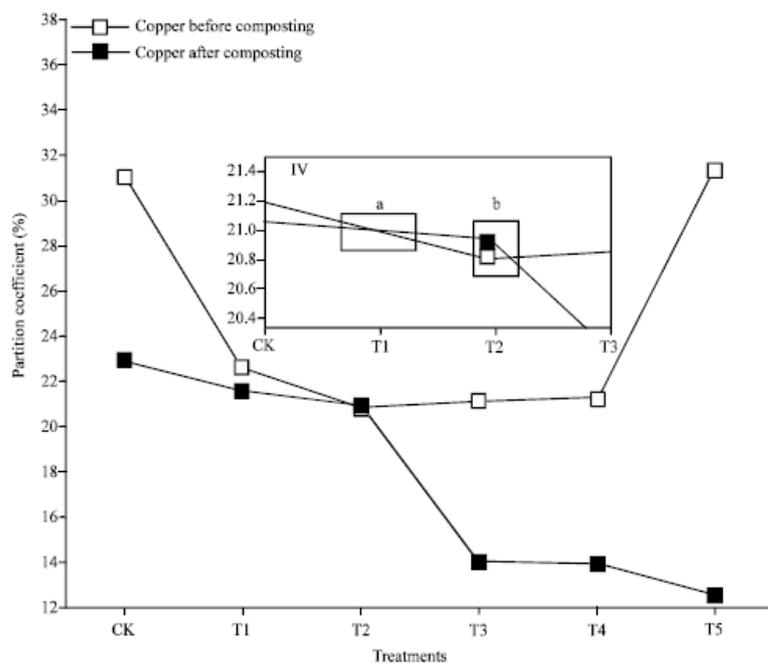


Fig. 3: Partition coefficient inflection point IV of heavy metal copper in cow dung samples before and after wood vinegar processing composting

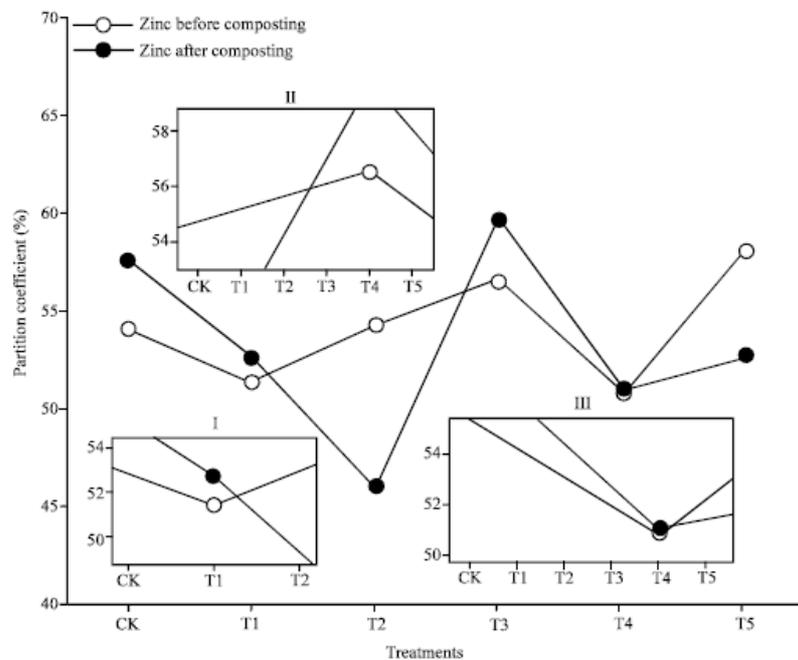


Fig. 4: Partition coefficient inflectionpoint I, II and III of heavy metal zinc in cow dung samples before and after wood vinegar processing composting

Table 5: The changes of the total concentration of heavy metals Zinc and DTPA extractable in cow dung samples before and after wood vinegar processing composting

Treatments:	Total concentration (mg kg^{-1})	DTPA-Copper concentration (mg kg^{-1})	Partition coefficient (%)	Partition coefficient difference (%)
CK				
Before composting	32.00±0.1	17.30±0	54.06	
After composting	45.23±0.12	26.07±0.06	57.63	-3.56 ^a
T1				
Before composting	36.00±0.2	18.50±0	51.39	
After composting	39.90±0.1	21.00±0	52.63	-1.24 ^a
T2				
Before composting	31.33±0.15	17.00±0	54.26	
After composting	49.20±0.1	22.63±0.06	46.00	8.25 ^a
T3				
Before composting	32.43±0.15	18.33±0.06	56.53	
After composting	33.17±0.06	19.80±0	59.70	-3.17 ^a
T4				
Before composting	36.83±0.12	18.70±0	50.77	
After composting	39.40±0.1	20.10±0	51.02	-0.25 ^a
T5				
Before composting	33.40±0.1	19.40±0	58.08	
After composting	39.10±0.1	20.60±0	52.69	5.40 ^b

Data is mean of 3 replicates±Standard Deviation. The negative number of the partition coefficient difference indicated: The extractable content increased in the proportion of the total content after the composting, the positive number indicated a decrease. In the same column the same letter indicated no significant differences between treatments at $p = 0.05$

This might attribute to "Concentration effect" significantly affects the total Copper concentrations and DTPA extractable concentrations, in addition, sawdust and added distilled water might bring about the "Dilution effect" (Huang, 2010) and the large formation of humus had chelation fixation effect on DTPA concentration which might reduce the **bioavailability** as well as enhance the "Concentration effect" in the late composting (Canarutto et al., 1991; Planquart et al., 1999).

Heavy metal zinc analysis: Ditto similar analysis, taken wood vinegar to handle the heavy metal zinc, (Table 5). The analysis results were shown in Table 6.

Table 6: The significant analysis of different treatments

Treatments:	Treatments:	Mean differences significance test
T2	CK, T1, T3, T4 and T5	Significant
T5	CK, T1, T3 and T4	Significant
T4	CK, T1 and T3	Significant
T1	CK and T3	Significant
T3	CK	Not significant

Similarly, these results demonstrated convincingly that using wood vinegar treatment the heavy metal zinc got good results. The passivation effect of treatment group was better than the control group. But, the passivation effect was proportional with increasing the content of wood vinegar, the passivation effect appeared that first increased, then decreased and then increased again, Experimental results showed that the wood vinegar has the best passivation effect for zinc there may be multiple proportion. That to add the proportion of zinc passivation effect in certain scope, the results showed that add proportion directly affect the passivation of zinc.

Experimental results showed that the partition coefficient of zinc appeared on the positive and negative alternating situations after composting (Table 5, Fig. 2 and 4). The partition coefficient had been reduced, respectively -3.56, -1.24, 8.25, -3.17, -0.25 and 5.40%. Compared with the control group, the partition coefficient of treatment groups was greater than the control group. Experimental results showed the wood vinegar had good passivation effect for zinc and the best passivation in treatment 5 was 8.25%. There had been a positive and negative situation in partition coefficient differences, showed the passivation effect of zinc under treatment of the wood vinegar

positive and negative oxidation in particular, consistent differences, showed the passivation effect of zinc under treatment of the wood vinegar. indicated instability, might be due to the combined effects a "Concentration effect", "Dilution effect" and "Chelation" were greater than the effect of wood vinegar treatment after composting (Wang et al., 2006). Therefore, the unstable of passivation was the result of comprehensive effects common cause in the composting process.

CONCLUSION

The activity and toxicity of heavy metals was not only related to biological effectiveness but also more closely related to chemical forms. Therefore, the activity and toxicity of water soluble states was the largest. Therefore, the changes in the form of heavy metals could accurately evaluating the effect of the **bioavailability** of heavy metals during the composting process.

Huang (2010) studies showed that the bamboo vinegar can reduce the **bioavailability** of Copper and Zinc in pig manure (Huang, 2010). He (2011) studied the transformation and **bioavailability** of heavy metals forms by adding passivator during composting process. The above results had provided the basis for the use of metal passivator during composting process. This study showed that, it could reduce its heavy metal **bioavailability** after wood vinegar deal with cow dung composting. The treatment method was important means and ways of reducing the risk of heavy metal pollutions. Due to the wood vinegar containing a variety of organic compounds, meanwhile heavy metals could be diluted and neutralized by wood vinegar. And the wood vinegar could also promote the transformation of heavy metal forms and reduce the harmful levels of heavy metals. Therefore, the additive proportion of the wood vinegar had a greater impact on the treatment of heavy metals. The passivation effect of heavy metal copper was 8.15% in the control group, second only to the maximum of 18.78% in the treatment group and higher than the other groups. The wood vinegar content should not be less than 0.80% for the processing of copper, however, for the treatment of zinc, the wood vinegar content at 0.35% was the best. In order to prevent the expansion of its fluctuation change influence in composting process, the stability of the passivation effects should be remained. Which means in order to prevent the effect of secondary factors was larger than the passivation for the wood vinegar after composting. The volume of the pile and the wood vinegar gradient should be increased at the same time, also the cycle of composting should be extended.

In actual production and application, in addition to consider the treatment effects, the raw materials sources of passivator also should be considered. Since the wood vinegar was obtained by agriculture and forestry residues after thermal cracking. Thinking from the aspect of the source, our country is rich in agricultural and forestry residues. Putting the wood vinegar as passivator can not only turn waste into treasure but also can recycling use of the waste of resources, realizing the harmless using of agricultural and forestry residues and the excrements of livestock in the end. It has the vital significance to the sustainable development of ecological agriculture. It is practical and feasible for choosing the wood vinegar as passivator.

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