Waste Management xxx (2017) xxx–xxx

Contents lists available at ScienceDirect

Waste Management

journal homepage: www.elsevier.com/locate/wasman

Seed germination test for toxicity evaluation of compost: Its roles, problems and prospects

Yuan Luo[a,b], Jie Liang[a,b,*], Guangming Zeng[a,b,*], Ming Chen[a,b], Dan Mo[a,b], Guoxue Li[c,d], Difang Zhang[c,d]

[a] College of Environmental Science and Engineering, Hunan University, Changsha 410082, China
[b] Key Laboratory of Environmental Biology and Pollution Control (Hunan University), Ministry of Education, Changsha 410082, China
[c] College of Resource and Environmental Science, China Agricultural University, Beijing 100193, China
[d] Beijing Key Laboratory of Farmland Pollution Prevention-control and Remediation, China

ARTICLE INFO

Article history:
Received 6 April 2017
Revised 18 September 2017
Accepted 19 September 2017
Available online xxxx

Keywords:
Compost
Toxicity
Germination
Model seed

ABSTRACT

Compost is commonly used for the growth of plants and the remediation of environmental pollution. It is important to evaluate the quality of compost and seed germination test is a powerful tool to examine the toxicity of compost, which is the most important aspect of the quality. Now the test is widely adopted, but the main problem is that the test results vary with different methods and seed species, which limits the development and application of it. The standardization of methods and the modelization of seeds can contribute to solving the problem. Additionally, according to the probabilistic theory of seed germination, the error caused by the analysis and judgment methods of the test results can be reduced. Here, we reviewed the roles, problems and prospects of the seed germination test in the studies of compost.

Contents

1. The roles of the seed germination test for the toxicity evaluation of compost

Composting, an effective and economical biotechnology, is widely used in the sanitization and recycling of biowastes including animal manures (Ge et al., 2016; Liang et al., 2017a) and carcasses (Gwyther et al., 2011), crop straws (Zhang et al., 2016), agro-industrial residues (Aviani et al., 2010), municipal organic solid wastes (Kelessidis and Stasinakis (2012); Anand and Apul, 2014), etc. Compost is often used as the growth media (Boldrin et al., 2010), the organic fertilizer (Feng et al., 2016), the soil amendment (D Hose et al., 2014; Alvarenga et al., 2015) and the suppressive substance of soil-borne plant diseases (Mehta et al., 2014; Yu et al., 2015) for agricultural production and landscaping. In recent years composting has been studied for the bioremediation of soils contaminated with heavy metal(loid)s (Park et al., 2011; Wu et al., 2016; Liang et al., 2017 b) and organic

* Corresponding authors at: College of Environmental Science and Engineering, Hunan University, Changsha 410082, China.
E-mail addresses: liangjie@hnu.edu.cn, liangjie82@163.com (J. Liang), zgming@hnu.edu.cn (G. Zeng).

https://doi.org/10.1016/j.wasman.2017.09.023
0956-053X/© 2017 Published by Elsevier Ltd.

contaminants (Chen et al., 2015; Huang et al., 2016; Kastner and Miltner, 2016).

Compost quality, such as stability and maturity, should be checked before the application of compost on land. The unstable and/or immature compost can have adverse effects on seed germination, plant growth and soil environment due to the decreased supply of oxygen and/or available nitrogen or the presence of phytotoxic compounds (Bernal et al., 2009). Stability is the resistance of the organic matter in compost against further microbial decomposition as long as there is no inhibition on the microbes by other factors not relevant to the organic matter, while maturity is an agronomic parameter that is clearly associated to the effect of compost on plant growth (Komilis, 2015). The indices of respiration and humification of compost are used to evaluate the stability and maturity of compost (Komilis and Kanellos, 2012; Hill et al. (2013); Nikaeen et al., 2015), respectively. The substances, including low molecular weight organic acids (e.g. phenolic acids (Marambe et al., 1992)), ammonium nitrogen (NH_4^+-N) (Ramírez et al., 2008), salinity (Hase and Kawamura, 2012), heavy metals (Fuentes et al., 2004), xenobiotics (e.g. antibiotics (Liu et al., 2009) and agrochemicals (Cayuela et al., 2008; Tang et al., 2008)), can cause damage to plants when they are in high levels. In general, many of these substances need to be gauged via time-consuming and expensive detection processes to determine whether their levels are beyond or within acceptable ranges. However, there exists the possibility for the unexpected factors that are not taken for analysis. Furthermore, there is a lack of analytical procedure to evaluate the joint effect of the toxic substances in compost. Consequently, as a bioassay, seed germination test has attracted a lot of attention to overcome these concerns. The seed germination index (*GI*) was firstly proposed by Zucconi et al. (1981) who used cress seeds in the germination test for evaluating the toxicity of compost. *GI* is calculated by the radicle length and germination percentage of the seeds in the sample (compost extract) compared to that in the control (e.g. deionized water). *GI* is correlated with some other biological and chemical indices for evaluating compost quality. The study of El Fels et al. (2016) showed that *GI* was positively correlated with the biological index of the *Artemia salina* cytotoxicity test for evaluating the toxicity of compost. In addition, *GI* is positively correlated with humification parameters (Gavilanes-Terán et al., 2016) while negatively correlated with the content of NH_4^+ (Tiquia et al., 1996; Guo et al., 2012). Therefore, the seed germination test has been broadly accepted for evaluating compost quality. In Italy, *GI* is listed in the quality assessment regulation of compost for commercialization (Cesaro et al., 2015). However, there are large variations in the aspects of method and seed species of the test among previous studies (see Section 2). The problems and prospects of studies in the seed germination test will be comprehensively reviewed in the following parts.

2. Could there be a widely accepted procedure of the seed germination test?

The seed germination test procedure consists of three major steps. Firstly, prepare an aqueous extract of compost; secondly, incubate seeds with the extract; thirdly, measure and calculate the indicators related to the test results by Eqs. (1–6), including the seed germination (*SG*), the relative seed germination (*RSG*), the relative radicle growth (*RRG*) and the seed germination index (*GI*). More details of the typical test procedures in studies are summarized in Table 1.

$$SG = \frac{\text{Number of germinated seeds}}{\text{Number of total seeds}} \times 100\% \quad (1)$$

$$RSG = \frac{\text{Number of germinated seeds(sample)}}{\text{Number of germinated seeds(control)}} \times 100\% \quad (2)$$

$$RRG = \frac{\text{Total radicle length of germinated seeds(sample)}}{\text{Total radicle length of germinated seeds(control)}} \times 100\% \quad (3)$$

$$GI = RSG \times RRG \times 100\% \quad (4)$$

$$GI = \frac{GI_{50\%} + GI_{75\%}}{2} \times 100\% \quad (5)$$

$$GI = \frac{GI_{25\%} + GI_{50\%} + GI_{75\%}}{3} \times 100\% \quad (6)$$

where $GI_{25\%}$, $GI_{50\%}$ and $GI_{75\%}$ were *GI* values of the samples that were 25%, 50% and 75% of the raw aqueous extract of compost diluted with deionized or distilled water (v/v), respectively.

At present, the major problem in the studies of the seed germination test is that there is no universally adopted procedure, which is reflected at the incompleteness of the test procedure and the differences of the corresponding methods (Table 1). In the first step, it is essential to determine the extraction ratio because the toxicity of compost extract is related to its concentration (Emino and Warman, 2004; Said-pullicino et al., 2007; Young et al., 2016). Although the extraction ratio of 1:10 (w/v) is one of the most used ratios in studies (Table 1), the test samples are fresh samples and have different moisture content. Owing to the large change in the moisture content of the raw material during composting, the moisture content of the samples collected from different composting stages is different (Chikae et al., 2006). In order to accurately evaluate the change of the toxicity of samples during composting and increase the comparability of the toxicity of samples with different moisture content, it is necessary to eliminate the interference of moisture content on test results. So far, there are two methods to address this problem. One method (Chikae et al., 2006; Khan et al., 2014) is to prepare the extract according to a certain extraction ratio of water and dry weight of fresh sample, but how to select a suitable value of the extraction ratio remains unsettled. Another method (Pampuro et al., 2010) is to adjust the moisture content of fresh sample to 85% to prepare the extract or its dilutions for the seed germination test (Said-pullicino et al., 2007; Cesaro et al., 2015). Centrifugation and membrane filtration can quickly and effectively remove the particulates in the extract. They are essential steps to reduce the interference of particulates on test results. In the second step, the major differences are reflected at the species of seed and the definition of seed germination (Table 1). There is no recognized seed species that can be used to evaluate the toxicity of compost and the seed is often obtained locally. The definition of seed germination can be divided into three groups by radicle length: only visible, at least 2 mm and at least 5 mm. It is not difficult to observe, even the incubation time is prolonged, the initial radicles of the germinated seeds inhibited in test are no longer to elongate, so a certain length that the radicle reached is used as the operational definition of germination. In fact, the process of germination is completed by visible radicle protrusion through the testa (seed coat), and radicle elongation belongs to post-germination (Weitbrecht et al., 2011). Moreover, if the incubation time is too long, the secondary root will begin to grow and the cotyledon will have strong phototaxis, and the length of radicle cannot exactly reflect the toxicity. The incubation time depends on specific environmental conditions and seed species. After the incubation time is over, seeds can be frozen at -10 °C for 24 h or added with ethanol equivalent to the sample (aqueous extract) to end their growth (Macias et al., 2000; Gómez-Brandón et al., 2008). These methods are effective in the handling of seeds,

Please cite this article in press as: Luo, Y., et al. Seed germination test for toxicity evaluation of compost: Its roles, problems and prospects. Waste Management (2017), https://doi.org/10.1016/j.wasman.2017.09.023