ORGANIC MATERIAL DECOMPOSITION CAPACITY OF INDIGENOUS MICROORGANISM COMMUNITIES FROM DIFFERENT FARMING SYSTEMS IN SOC TRANG PROVINCE, VIETNAM

Le Thi Xa[1], Nguyen Khoi Nghia[2*], Hüseyin Barış Tecimen[3]

[1] School of Education, Soc Trang Community College
[2] Department of Soil Science, College of Agriculture, Can Tho University
[3] Department of Soil Science and Ecology, Faculty of Forestry, Istanbul University-Cerrahpaşa, Istanbul-Turkey

SUMMARY

This study aimed to assess fifteen indigenous microorganisms (IMOs) ability to decompose the rice straw, sugarcane bagasse, coco peat, sawdust, and rice husk as organic materials collected from different farming systems in Soc Trang province, Vietnam. Microbial density capable for cellulose decomposition was determined by the plate counting method on agar medium containing 1% carboxymethylcellulose. Cellulose degradation capacity of IMOs was qualified by Congo red method with determination of a halo zone diameter. Finally, the decomposition capacity of IMOs for organic materials was determined by the mass loss method after 30 days of inoculation under laboratory conditions. The results showed that the density of cellulose decomposer in IMOs was around 10^5 CFU/g and all 15 IMOs had halo zone diameters varying between 3.38 - 8.63 cm. Results of the decomposition experiment for five different organic materials under the laboratory conditions revealed that percentage of decomposition of rice straw material was ranged between 52.2 - 57.9%. The percentage of sugarcane bagasse decomposition varied from 19.03 to 47.72% while the values for the control treatments without IMOs inoculation of these two materials were 20.39% and 11.81%, respectively. However, for other materials including coco peat, sawdust and rice husk, all fifteen IMOs exhibited very low capacity in decomposition and there was no significant difference between the treatments with and without IMOs inoculation regarding to the decomposition rate of the organic materials. The results of this study indicated that IMOs can be used to enhance the decomposition rate of some agricultural waste products in the composting process.

Keywords: Organic decomposition, indigenous microorganism community, organic materials, rice straw, sugarcane bagasse.

INTRODUCTION

Vietnam is facing with the utilization of agricultural harvest residues. The amount of discharged rice straw and rice husk annually reached approximately ten million tons during the process of harvesting and processing rice, whereas, recycling or re-evaluation of sawdust, cocopeat, and sugarcane still requires a solution (Cam *et al*., 2015). The decomposition of cellulose, hemicellulose, and lignin-rich wastes is complex and tardily. The decomposition of cellulose and lignin by physical and chemical methods is complex, costly, and environmentally toxic, treating cellulose and lignin-containing organic wastes with microorganisms incorporated processes are efficient and eco-friendly (Cam *et al*., 2015).

The microbes play a pivotal role in the decomposition of calcitrant and complex organic compounds into elemental and available forms. The natural soils are the abode of microorganisms capable of degrading lignocelluloses material of dead plants. Fungal species are predominately degrading cellulose and lignin into simple sugars and phenolic acids (Man and Ha, 2006), which subsequently provided to other microbes in the soil. Some of the soil fungi economically employed in crop soil amendments are species of *Trichoderma* and *Pleurotus* (Jayadeva *et al*., 2010).

Organic resources play an essential role in soil fertility management in the tropics by their short-term effects on nutrient supply and longer-term contribution to soil organic matter (SOM) formation (Palm *et al*., 2001). The decomposition rate and the amount of nutrient release from organic matter (OM) determine the short-term benefits of tree residues for crop nutrition (Handayanto *et al*., 1997). To design more efficient agricultural systems, there should be a clear understanding of the determinants of nutrient supply, especially those that reduce the time of organic material decomposition and byproducts releasing from agricultural residues.

Plenty of beneficial microbes are included in indigenous microorganisms (IMOs) and, they bear an implicit potential of decomposer agent for making compost. The IMOs are also sprayed over rice straw before the rice field undergoes a tilling process to promote the decomposition of rice straw as well as other organic materials.

The beneficial microorganism inoculation helped to improve the efficiency of the degradation process (Biey *et al*. 2000). Degradation of organic compound by IMOs without any artificial enhancement is termed as “intrinsic bioremediation” and this is one of the best remedial actions applied without any soil contamination. Addition of IMOs into compost helps to facilitate the decomposition process. Indigenous microorganisms include filamentous fungi, yeasts and bacteria collected from noncultivated soil. It has a high amount of microorganisms on the soil and often found under bamboo trees (Abu-Bakar and Ibrahim, 2013) as well as other cultivation fields like crop rotation, banana, shallot, vegetables, rice, watermelon, grassland, maize, lettuce, oranges, grapefruit, guava and sugarcane (Xa *et al*., 2018). Since microbial community plays an important role in organic material decomposition. However, the study about using the IMOs to enhance the organic material decomposition has limited. Therefore, the aim of this study was to assess the ability for organic material decomposition of fifteen IMOs from different farming systems in Soc Trang province, Vietnam.

MATERIAL AND METHOD

Materials

Fifteen different IMOs were collected from different crop systems in Soc Trang province, Vietnam including bamboo, crop rotation (corn-watermelon-courgette), banana, shallot, vegetables, rice, watermelon, grassland, maize, lettuce, oranges, grapefruit, guava, sugarcane. Refer to Xa and Nghia (2019) for more information about the origin of sampling sites.

Cell counting of microbes in collected IMOs capable in decomposing cellulose

An aliquot of 10 grams of each IMOs was put into a 250 mL glass bottle containing 90 mL sterilized distilled water on a shaker at a speed of 150 rpm for one hour and was maintained for 5 minutes after shaking. Prepared a series dilution with a factor of 10 containing, aliquots of 50 µL of each dilution were spread on carboxymethylcellulose (CMC) agar plates. Each dilution for each medium was repeated within three replicates. The composition of 1-liter CMC medium containing 1 g $(NH_4)_2SO_4$, 1 g K_2HPO_4, 0.5 g $MgSO_4$, 0.001 g NaCl, 10 g CMC and 15 g agar (pH 7) was used for the IMOs inoculation (Ulrich *et al*., 2008). Samples were placed in incubators at 30^oC for 2 days. Finally, the plates were counted the number of colony-forming units (CFU) developed on agar medium to calculate the number of microbes.

Determination of cellulose degradation capacity of IMOs

Cellulose degradation capacity of IMOs was qualified by Congo red method with a halo zone diameter determination. An aliquot of 10 grams of each IMOs was put into a 250 mL glass bottle containing 90 mL sterilized distilled water on a shaker at a speed of 150 rpm for an hour and left stand for 5 minutes after shaking. An aliquot of 10 µL IMOs dilution was dropped on the central of CMC agar plates and left stand for 30 minutes. The plates were incubated at room temperature (30°C) for three days to allow microorganisms growth. After incubation, 10 mL of 0.5% Congo red staining solution was added to the plate and was shaken for 15 minutes. The Congo red staining solution was discarded and added 10ml of 1M NaCl to destain the plates by shaking for 15 minutes. Finally, 1M NaCl was discarded and the stained plates were analyzed by observing the formation of clear zone (halo zone) around the IMOs growth. The halo zone diameter was determined by the average perpendicular diameter of the halo zone.

Evaluation of decomposition capacity of IMOs for organic materials

Decomposition capacity of IMOs for organic materials was determined by the mass loss method after 30 days of inoculation under laboratory conditions (Cam *et al*., 2015). Five different organic materials subjected to decomposition in this study were rice straw, rice husk, sugarcane bagasse, coco peat and sawdust. Rice straw and sugarcane bagasse were cut into 2 cm-short pieces. All organic materials were washed with distilled water, gently spread and allowed to dry in laboratory conditions. To incubate the organic materials, 20 g (dry weight) of rice straw, rice husk, sugarcane bagasse, coco peat and sawdust was put into a 250 g round plastic box and added 2 ml of IMOs solution to a final density of 10^7 CFU/g and then added water to a moisture content of 80%, mixed well and left to stand in laboratory conditions for 30 days. Finally, the percentage of decomposition was calculated based on the residue dry mass.

Decomposition rate (%) = (initial dry mass - residual dry mass)*100/initial dry mass

Data analysis

The data were analyzed by ANOVA by MINITAB version 16.2 software.

RESULTS AND DISCUSSION

The number of microbes in collected IMOs capable in decomposing cellulose

The results of cellulose decomposing microbial counts showed no significant difference between IMOs crop systems. Microbial numbers including bacteria and fungi in the IMOs ranged from 1.80×10^5 to 6.53×10^5 CFU/g IMOs (Table 1). The highest bacterial CFU count observed in IMOs collected from vegetable grove soil and, secondly, the IMOs collected from grassland field had the number of microbes 6.13 x 10^5 CFU/g IMOs. The lowest number of bacteria was found in IMOs collected from sugarcane soil, with a number of 1.8 x 10^5 CFU/g IMOs, while the remaining number of IMOs microbes including bamboo, crop rotation, banana, shallot, rice, watermelon, maize, lettuce, oranges, grapefruit and guava varied from 2.27 x 10^5 to 5.33 x 10^5 CFU/g IMOs.

Table 1. The number of decomposing cellulose microorganisms

Number	Code	Origin of samples	Number of microorganisms (10^5 CFU/g)
1	IMO1	Bamboo	5.07
2	IMO2	Crop rotation	3.00
3	IMO3	Banana	2.27
4	IMO4	Shallot	3.40
5	IMO5	lettuce	5.13
6	IMO6	Rice	4.67
7	IMO7	Watermelon	3.80
8	IMO8	Grassland	6.13
9	IMO9	Maize	3.00
10	IMO10	Vegetables	6.53
11	IMO11	Oranges	2.80
12	IMO12	Grapefruit	2.60
13	IMO13	Guava	5.33
14	IMO14	Sugarcane	1.80
15	IMO15	Mix	5.40

**Note: Values in the same column with the same letters are not significant difference ($p<0.05$)*

This result clearly revealed that there was a presence of high numbers of microbial cellulose decomposition in all fifteen studied IMOs and this result implies that almost all IMOs could be considered as a good source of beneficial microbes for soil improvement as well as plant growth promotion (Reddy, 2011). They can survive better under the extreme climatic conditions of the local environment than under artificial cultures and environments. Since they have resided and already adapted with the local conditions, they are considered as the best survival source of microbes for soil and plant improvement effectively.

Determination of cellulose degradation capacity of IMOs

The results indicated that the halo diameters varied widely among IMOs systems (Figure 1) and their cellulose degradation capacity was significantly different among each other ($p<0.05$) (Table 2). A tested IMOs from guava cultivation fields created the highest halo zone (8.63 cm), closely followed by IMOs collected from vegetables and shallot systems fields 7.70 and 7.47 cm, respectively which are significantly higher than other IMOs. Meanwhile, the remaining IMOs including bamboo, crop rotation, banana, lettuce, rice, watermelon, grassland, maize, oranges, grapefruit and sugarcane exhibited halo zone formations varying from 3.38 to 6.23 cm. It was noteworthy that when mixing partly all the collected IMOs together to have an integrated IMOs, the average diameter of the halo zone produced by this microbial community significantly increased. Therefore, a combination of several IMOs from different ecosystem habitats is another approach and is very essential to have better function of IMOs (Reddy, 2011; Xa *et al*., 2018).

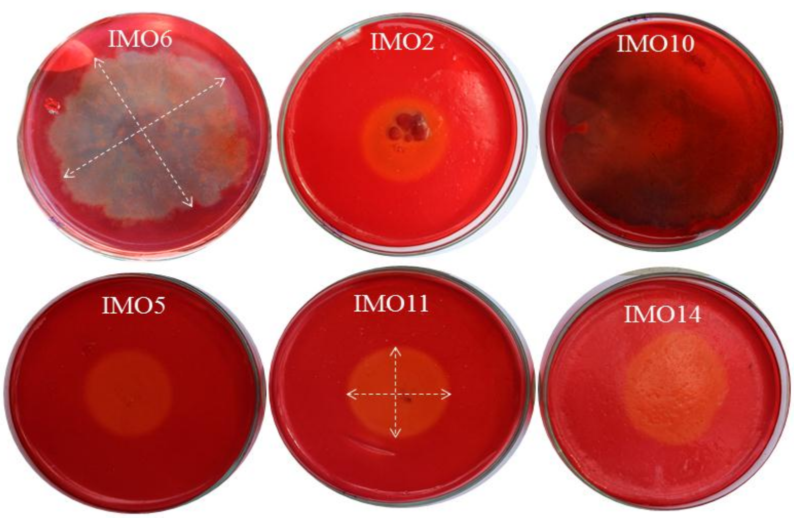

Figure 1. The variety cellulose resolution halo zone diameters of some indigenous microorganism

Table 2. Diameter size of cellulose resolution halo of 15 IMOs

Number	Code	Origin of samples	Halo zone diameter size (cm)
1	IMO1	Bamboo	3.77[e]
2	IMO2	Crop rotation	3.50[e]
3	IMO3	Banana	4.03[e]
4	IMO4	Shallot	7.47[b]
5	IMO5	lettuce	3.43[e]
6	IMO6	Rice	6.23[cd]
7	IMO7	Watermelon	4.20[e]
8	IMO8	Grassland	3.38[e]
9	IMO9	Maize	3.80[e]
10	IMO10	Vegetables	7.70[ab]
11	IMO11	Oranges	3.76[e]
12	IMO12	Grapefruit	3.50[e]
13	IMO13	Guava	8.63[a]
14	IMO14	Sugarcane	5.57[d]
15	IMO15	Mix	6.87[bc]

** Note: Values in the same column with the same letters are not significant difference ($p < 0.05$).*

Decomposition capacity of IMOs for organic materials

Rice straw

The decomposition capacity of 15 IMOs for rice straw results after 30 days of inoculation showed that all IMOs have significantly higher decomposition rates compared to ($p < 0.05$) control treatments. The rice straw decomposition rates of 15 IMOs origins ranged from 52.2% to 57.9%, with no significant difference among each other.

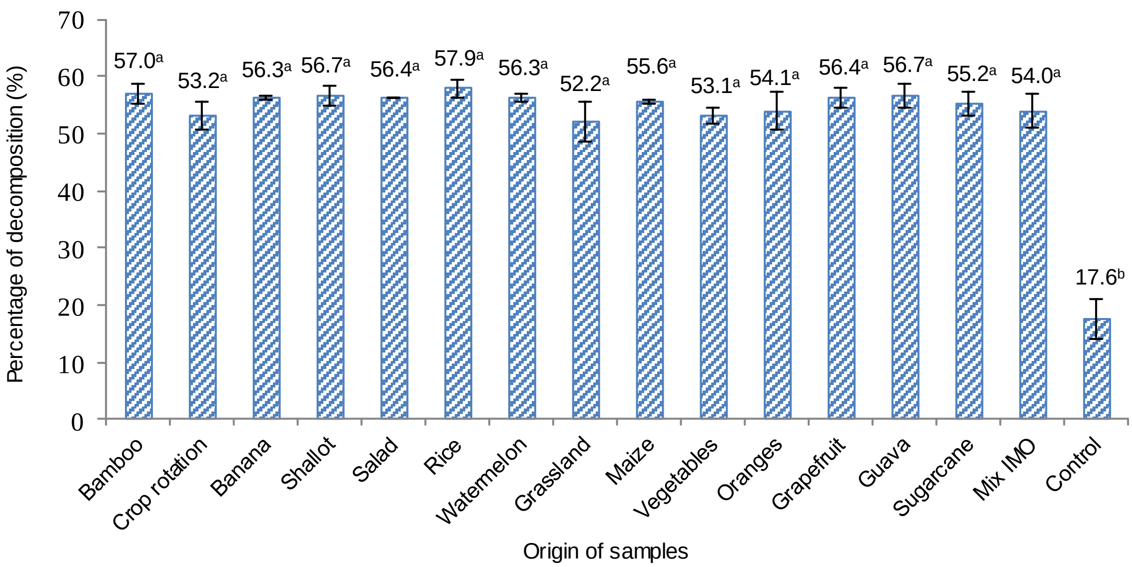

Figure 2. Percentage of rice straw material decomposition of 15 IMOs from different farming systems in Soc Trang province, Vietnam

We recorded that rice straw treated with microbial consortium exhibited better decomposition compared with rice straw treated with water - control in this experiment. This has confirmed more effective and efficient biological processes and enzymatic activities exerted by synergistically working microorganisms introduced to activate in a group of a consortium where their performance is higher than microorganisms on an individual population basis (Kato *et al*. 2005). Similarly, Tuerson *et al*. (2013) reported more stable, efficient, and successful microbial activities during their synergetic collaboration. Microorganisms in a consortium also maintain metabolic and ecological compatibility and stability. Wongwilaiwalin *et al*. (2010) reported that the biomass degrading capability is based on the functional and structural stability of a microbial consortium. Some microorganisms are superior to others in degrading different component of rice straw namely cellulose, hemicellulose and lignin. Thus, the most efficient bacteria in cellulose, hemicellulose and lignin degradation were mixed together to function as a strong and competent microbial consortium. The previous study of Bakar *et al*., (2018) suggested that mixed culture of microorganisms used, could improve the decomposition process of organic materials. Several researchers had also reported that rice straw decomposition process could be accelerated by using mixed culture of microbial inoculant (Zhao *et al*., 2014; Mohamed *et al*., 2016).

Compared to previous study by Cam *et al*., (2015) who isolated 17 strains of fungi which had capable of decomposing rice straw, rice husk, sugarcane bagasse, coco peat and sawdust and showed that all are possible rice straw decomposition was very high, ranging from 37.1% to 47.6% and was significantly higher from control treatment in this study. These results clearly suggested that IMOs collected in this study are efficiently capable on decomposing rice straw.

Sugarcane bagasse

Results of the decomposition experiments for organic materials of sugarcane bagasse after 30 incubation days revealed that different IMOs had different decomposing capacities and significantly higher (p<0.05) than control treatment. In fact, the decomposition percentage of sugarcane bagasse material varied between 20.6% – 49.6%, while the values for the control treatments without IMOs inoculation of this material were 11.81%. The highest capacity found in tested IMOs came from maize cultivation field at a rate of 49.6%, closely followed by three IMOs collected from cultivation soil of grassland, orange and grapefruit with a decomposing rate of 42,0%, 41.4% and 41,2%, respectively. Surprisingly, the IMOs collected from sugarcane field had the lowest decomposing rate at 20.6%, whereas the remaining IMOs including bamboo, crop rotation, banana, shallot, lettuce, rice, watermelon, vegetables, guava and mixed IMOs had the decomposition percentage ranged from 22.7% to 36.6%.

Torkashwand *et al*., (2008) recorded approximately 2/3 loss of sugarcane bagasse (urea added treatment) at *Trichoderma* fungi inoculated experiment after 10 weeks of incubation. Similarly, Cam *et al*. (2015) obtained sugarcane decomposition rates ranging from 33.2 to 46.9% by the fungal isolates. Among those, the strains of PH-L3 had a higher percentage of sugarcane decomposition (32.8%) compared to other strains.

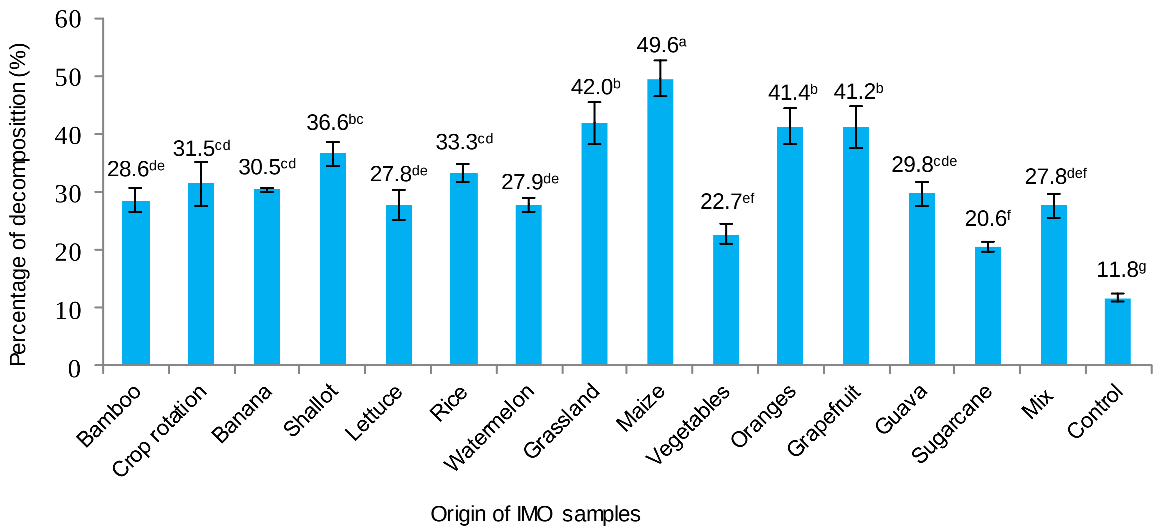

Figure 3. The percentage of decomposition of sugarcane bagasse material of fifteen indigenous microorganisms

Rice husk, coco peat and sawdust

Unlike the success of of IMOs in decomposing of rice straw and sugarcane, the results in decomposing of rice husk, coco peat and sawdust were very low. The immediately and freshly collected rice husk, coco peat, and sawdust materials from mills might have retarded and suppressed the decomposition. Moreover, these materials are difficult to decompose, and 30 days period would not be sufficient for microorganisms to degrade them.

Besides, Abu-Bakar and Ibrahim (2013) consider that addition of IMOs is not the main factor in determining the rate of degradation but the C/N ratio value of the compost could be an alternative factor that causes to raise in temperature, thus promotes the degradation. In addition, Cam *et al*. (2015) analyzed the components of rice straw, sugarcane, rice husk, sawdust and cocopeat and showed that these substances contain high total carbon and subsequently high C/N ratio while total nitrogen, total potassium, total phosphorus are remarkably low (except for rice straw) which leads to retardant decomposition of these materials. Thus, we agree that the total values of N, P and K of rice straw, sawdust and coco peat are efficient on decomposition of those materials. Because composting rate depends on a great extent of C/N ratio, lignin and polyphenol contents, the presence or absence of suitable microbial agents of decomposition etc. (Wilson 1989; Prabhu and Thomas 2002). Therefore, these affects the decomposition speed of three materials like rice husk, coco peat and sawdust of the fifteen IMOs test.

CONCLUSIONS

The investigation results showed that fifteen different IMOs collected from different farming ecosystems in Soc Trang province contained microbial communities that were able to decompose lignocellulose containing materials, and synthesized cellulase enzyme involving in decomposition of organic materials. Furthermore, these fifteen IMOs also helped to boost decomposition rate of some organic materials, especially the rice straw and the sugarcane bagasse. The results impose that IMOs has great potential in the treatment of agricultural waste products and can be used to accelerate the decomposition of agricultural waste products in the composting process for organic fertilizer production to help to improve soil, to increase growth, yield, and quality of crops.

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KHẢ NĂNG PHÂN HUỶ VẬT LIỆU HỮU CƠ CỦA HỆ VI SINH VẬT BẢN ĐỊA THU THẬP TỪ CÁC HỆ THỐNG CANH TÁC CÂY TRỒNG KHÁC NHAU Ở TỈNH SÓC TRĂNG, VIỆT NAM

Lê Thị Xã[1], Nguyễn Khởi Nghĩa[2*], Hüseyin Barış Tecimen[3]

[1] *Khoa Sư phạm, Trường Cao đẳng Cộng đồng Sóc Trăng*
[2] *Bộ môn Khoa học Đất, Khoa Nông nghiệp, Trường Đại học Cần Thơ*
[3] *Bộ môn Đất và Sinh thái, Khoa Lâm Nghiệp, Trường Đại học Istanbul University-Cerrahpaşa, Thổ Nhĩ Kỳ*

TÓM TẮT

Mục tiêu của nghiên cứu này nhằm đánh giá khả phân hủy các vật liệu hữu cơ gồm rơm, xác mía, mụn dừa, mùn cưa và vỏ trấu của 15 hệ vi sinh vật bản địa (IMO) thu thập từ các hệ thống canh tác cây trồng khác nhau ở tỉnh Sóc Trăng, Việt Nam. Mật độ vi sinh vật phân huỷ cellulose được xác định bằng phương pháp đếm số khuẩn lạc trên môi trường agar có bổ sung 1% carboxymethylcellulose. Khả năng phân hủy cellulose của các IMOs được xác định bằng phương pháp xác định đường kính vòng phân giải phản ứng với thuốc thử Congo red. Cuối cùng, khả năng phân hủy đối với các vật liệu hữu cơ của các IMOs được xác định bằng phương pháp tính phần trăm khối lượng chất khô giảm đi sau 30 ngày dưới điều kiện phòng thí nghiệm. Kết quả cho thấy mật độ vi sinh vật có khả năng phân hủy cellulose trong các hệ IMOs tương đương 10^5 CFU/g và tất cả 15 IMOs đều có đường kính vòng phân giải thay đổi trong khoảng từ 3,38 đến 8,63 cm. Kết quả thí nghiệm phân hủy 5 loại vật liệu hữu cơ khác nhau trong điều kiện phòng thí nghiệm cho thấy tỷ lệ phân hủy rơm rạ dao động trong khoảng từ 52,2 đến 57,9%. Tỷ lệ phân hủy bã mía dao động từ 19,03 đến 47,72% trong khi giá trị ở nghiệm thức đối chứng không chủng IMOs của hai nguyên liệu này lần lượt là 20,39% và 11,81%. Tuy nhiên, đối với các vật liệu còn lại gồm mụn dừa, mùn cưa và trấu, tất cả 15 hệ IMOs đều thể hiện khả năng phân hủy rất thấp và không có sự khác biệt đáng kể giữa các nghiệm thức chủng và đối chứng không chủng IMOs về tốc độ phân hủy của các vật liệu hữu cơ. Kết quả của nghiên cứu này cho thấy các hệ vi sinh vật bản địa IMOs có thể được sử dụng để tăng cường tốc độ phân hủy của một số phế phẩm nông nghiệp.

Từ khóa: Hệ vi sinh vật bản địa, phân huỷ hữu cơ vật liệu hữu cơ, rơm, xác mía.

[*] Author for corresspondence: Tel: +84-932801727; Email: nknghia@ctu.edu.vn