Bamboo Powder as an Ingredient of Potting Substrates for Horticultural Plants

Takashi FUDANO¹⁾, Hiroyuki KIKUKAWA²⁾, Fumio KISHIDA³⁾

[Abstract]

As the spreading of abandoned bamboo forests is becoming a serious problem in Japan, new possible uses for bamboo biomass are being sought toward controlling their further spread. In the present study, we investigated the potential of bamboo powder substrates in cultivation of horticultural plants as well as for bamboo powder to be used as a soil supplement in potting substrate recycling. Twenty varieties of ornamental flowering species were sown either in a conventional potting substrate or bamboo powder substrates. After 30 days, the cumulative germination rate of each variety was not affected by the type of substrate the plants were grown in. Although the overall plant growth of pot French marigold, pansy, and strawberry showed a tendency of being inferior in the bamboo powder substrates compared with the conventional potting substrate, it was improved by the addition of compost. The growth of French marigold also improved when the fermented bamboo powder substrate was additionally fertilized. We found that the microbiota present in the recycled potting substrate containing fermented bamboo powder was similar to that found in fresh potting substrate. These results indicate that bamboo substrate has great potential to be employed both as a growing substrate for various garden plants and as a soil supplement in recycling of used potting substrate.

Key words : compost, fermented bamboo powder, microbiota, soil supplement in recycling

1. Introduction

The expansion of abandoned bamboo forests is a serious issue in Japan, particularly in the temperate regions, where abandoned Japanese timber bamboo (Phyllostachys bambusoides Sieb. et Zucc) and moso bamboo (P. edulis(Carrière)Houz.) are invading the surrounding thickets and artificial forests (Suzuki, 2008; Torii, 1998). In Japan, bamboo has been used since ancient times both as a food and in the production of goods that are indispensable for daily life (Torii, 2003). However, as lifestyle became more westernized and living standard increased after World War II, the quantity and variety of everyday bamboo products have greatly reduced. Consequently, the number of unmanaged bamboo groves has increased: although bamboo forests in Japan covered 148,000 ha in 1974, this area increased every year, growing to 159,000 ha by 2007, 161,000 ha by 2012, and 166,000 ha in 2019 (State of Forest Resources, Ministry of Agriculture, Forestry and Fisheries, http://www. rinya.maff.go.jp/j/keikaku/genkyou/ h24/pdf/ soukatsu_47_h24.pdf, 2021.2). Suzuki and Nakagoshi (2008) reported that moso bamboo forests expanded at approximately 2% per year in regions with reduced shipments of bamboo shoot harvests. The expansion of abandoned bamboo groves is proved to have negative environmental effects such as the degradation of rural scenery, reduced biodiversity, increased damage to agricultural crops by wild animals, and landslides. To control this expansion, new uses for bamboo biomass must be found.

In recent years, the area under floriculture and the overall quantity of flowering plants shipped considerably decreased (Ministry of Agriculture, Forestry and Fisheries, http://www.maff.go.jp/j/tokei/ kouhyou/sakumotu/sakkyou_kaki/attach/pdf/index-2. pdf, 2021.2). Comparing the shipping volumes for 2007

Graduate School of Landscape Design and Management, University of Hyogo/Hyogo Prefectural Awaji Landscape Planning & Horticulture Academy

²⁾ Hyogo Prefectural Agricultural High School

³⁾ Experimental Farm, Graduate School of Agriculture, Kyoto University

and 2016, the volume of cut flowers, pot plants, and seedlings for flower gardens decreased by 20%, and the volume of bulbs shipped decreased by over 40% during this period. To increase floricultural product consumption amid these circumstances, it is necessary to give products a new, original value.

It has reported that one factor that limits pot plant sales was the lack of ease with which the potting substrate can be disposed of when plants finish their life cycle (The Tokai Regional Agricultural Administration Office, https://www.maff.go.jp/tokai/ tokei/tyo-sei/ish-iko/pdf/20160129_hana.pdf, 2021.2). Many local governments prohibit the disposal of potting substrate as combustible garbage, and people can be found falsely passing it on as combustible garbage or illegally dumping it in parks or riverbeds. The bamboo used in the present study can be disposed of as combustible garbage, and we deem that, if potted flowering plants are grown in substrates containing bamboo, they would have an added value of being able to be disposed of as household combustible garbage and would be favored among consumers.

In the present study, to obtain information on how to produce bamboo-based high value-added pot plant products, we investigated the germination and growth of horticultural plants in substrates containing bamboo as well as the potential of using bamboo powder as a soil supplement.

2. Materials and Methods

2.1 Using Bamboo in Potting Substrates2.1.1 Seed Germination

In total, 20 varieties of ornamental flowering species were used for the germination experiments: Baby blue eyes (Nemophila menziesii), baby's breath (Gypsophila elegans), brompton stock (Matthiola incana), Canterbury bells (Campanula medium), China aster (Callistephus chinensis), cockscomb (Celosia argentea var. cristata), common zinnia (Zinnia violacea), cypress vine (Ipomoea quamoclit), English daisy (Bellis perennis), French marigold (Tagetes patula), garden balsam (Impatiens balsamina), garden cosmos (Cosmos bipinnatus), horned violet (Viola cornuta), Japanese morning glory (Ipomoea nil), pansy (Viola × wittrockiana), petunia (Petunia × hybrida), pot marigold (Calendula officinalis), Roger's bronze leaf (Rodgersia podophylla), scarlet sage (Salvia splendens), and sunflower (*Helianthus annuus*). On February 6, 2014, 10 seeds of each variety were sown into pots (diameter 9 cm) filled with bamboo powder or a commercial potting substrate (Pro-Mix PGX, Premier Tech Horticulture). This bamboo powder was purchased from a lumber company (Take no chikara, Sugimoto Corp., Hyogo) in a sealed packet. As this bamboo powder was found to be acidic (Fudano et al. 2016), 18 g of garden lime per liter of bamboo powder was added to obtain the neutral pH (6.3–6.8). There were two replicates per treatment. Seeds were germinated in a glasshouse at the Awaji Campus for Landscape Design and Management, University of Hyogo, and their germination rate was recorded over a 30-day period.

2.1.2 Pot Marigold and Pansy Cultivation

On November 29, 2013, we sowed pot marigold seeds (Calendula officinalis, cultivars: 'Orange Star', 'Gold Star', and 'Shinkuro') into 200-cell plug trays. Seedlings were transplanted into pots (diameter 9 cm) filled with potting substrate on February 4, 2014. Three different potting substrates were used as follows: (1) 7:3 blend of Akadama soil (red ball earth) + pH-adjusted peat moss, (2) 7:3 blend of Akadama soil (red ball earth) + bamboo powder, and (3) bamboo powder. The potting substrates were supplemented with 3 g of slow-release NPK (14:11:13) fertilizer and 1 g of fused phosphate fertilizer per liter. There were 10 replicates per treatment. The length and diameter of main stem, the number of leaves and SPAD were scored immediately, 2 and 4 weeks after transplantation. SPAD was measured using a chlorophyll meter (SPAD-502, MINOLTA). Four weeks after transplantation, the fresh weights of shoot and root were measured. Subsequently, the plant material was dried at 80°C for 3 days and the dry weights were obtained.

On September 3, 2013, pansy seeds (Viola × wittrockiana, cultivar: 'Nature Blue and Yellow') were sown into 200-cell plug trays filled with a commercial potting substrate (Pro-Mix PGX). On October 30, 2013, seedlings were transplanted into pots (diameter 9 cm) filled with potting substrate. Four different potting substrates were used: (1) commercial horticultural potting substrate, (2) bamboo powder, (3) 1:1 blend of bamboo powder and bamboo chips, and (4) 1:2:1 blend of bamboo powder, bamboo chips, and bark compost. The size of bamboo chips was approximately 3 cm \times 2 cm \times depth 1 cm,

and these were provided by Sugimoto Corp. On December 3, 2013, the seedlings were potted into pots (diameter 15 cm) filled with the potting substrates mentioned above. There were five replicates per treatment. Plant height, spread, and SPAD were measured 6, 8, 10, and 12 weeks after transplantation. Black and white image analysis software (Adobe Photoshop CS6, Adobe) was used to analyze photographs taken from 50 cm above the plants' top to measure plant spread, and the results were shown as pixel counts. Twelve weeks after transplantation, the number of open flowers was counted on each plant.

The pot marigold and pansy plants were cultivated in a glasshouse at the Awaji Campus for Landscape Design and Management, University of Hyogo.

2.1.3 Growing Strawberries

On December 5, 2013, strawberry (Fragaria × ananassa, cultivar: 'Sagahonoka') seedlings were transplanted into pots (diameter 15 cm) and filled with one of the following four types of potting substrate: (1) 2:1 blend of peat moss and vermiculite (conventional potting substrate), (2) bamboo powder, (3) 1:1 blend of bamboo powder and bamboo chips, or (4) 1:2:1 blend of bamboo powder, bamboo chips, and bark compost. The potting substrates were supplemented with 5 g of garden lime, 5 g of fused phosphate fertilizer, and 12 g of slow-release NPK (14:11:13) fertilizer per liter. After transplantation of the strawberry seedlings, the pots were placed on trays and supplied with tap water. There were five replicates per treatment. The plants were cultivated in a plastic greenhouse at the Experimental Farm of Graduate School of Agriculture, Kyoto University. After 8, 10, 12, 14, 16, and 18 weeks, the length and diameter of the third leaf petiole; length, width, and SPAD of the highest open leaves; number of leaves; and diameter of the crown were measured for each plant. The weight and Brix of fruits harvested up to 18 weeks after transplantation were scored. Brix was measured using a Brix meter (Pocket refractometer PAL-J, Japan).

2.2 Substrate Recycling Using Bamboo Powder 2.2.1 Plant Cultivation in Recycled Substrates

We used the following eight types of potting substrates: (1) unused commercial potting substrate (Pro-Mix PGX; new substrate), (2) Pro-Mix PGX potting substrate previously used for growing cyclamens (used substrate), (3) used substrate that had been covered by black plastic for 10 days

(solarized substrate), (4) used substrate that had been steam-sterilized at 100°C for two hours (steamsterilized substrate), (5) used substrate mixed with 30% (v/v) fermented bamboo powder (fermented bamboo powder substrate), (6) used substrate mixed with 30% (v/v) unfermented bamboo powder, (unfermented bamboo powder substrate), (7) fermented bamboo powder substrate supplemented with 3 g per liter of slow-release NPK (10:10:10) fertilizer (fermented bamboo powder substrate with fertilizer), or (8) unfermented bamboo powder substrate supplemented with 3 g per liter of slowrelease NPK (10:10:10) fertilizer (unfermented bamboo powder substrate with fertilizer). It has previously been shown that a highly concentrated lactic acid bacterium is associated with bamboo powder (Otani and Iwasaki, 2007). (5) and (7) were bamboo powders purchased from a lumber company, whereas (6) and (8) were aerated bamboo powders purchased from a lumber company. On June 4, 2018, French marigolds 'Safari Yellow' were sown in trays filled with the potting substrates (1) to (6) described above, and on June 13, the seedlings were transplanted to pots (diameter 9 cm) filled with the potting substrates (1)to (8). Seedlings germinated in the substrates (5) and (7) were transplanted into pots filled with the potting substrate (5) or (7), and seedlings germinated in the substrate (6) were transplanted into pots filled with the potting substrate (6) or (8) and further cultivated in a greenhouse at Hyogo Prefectural Agricultural High School. There were five replicates per treatment. We recorded the germination rate from day 2 to 8 for each type of potting substrate, as well as the length of the main stem and the number of leaves at 1, 2, 3, 4, and 5 weeks after transplantation.

2.2.2 Recycled Substrate Microbiota

On June 5, 2018, we sent samples of the potting substrates (1) to (6) described above for amplicon analysis at a specialized institution to obtain a comprehensive understanding of the microbiota in each type of substrate.

Results

3.1 Using Bamboo in Potting Substrates

3.1.1 Seed Germination

Japanese morning glory, pansy, and scarlet sage showed a tendency to have a higher germination rate in

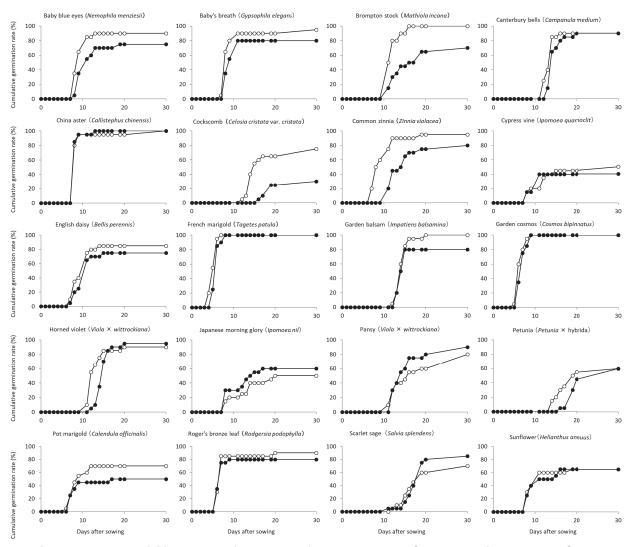


Fig. 1 Seed germination of 20 varieties of ornamental flowering species. ○: Pro-Mix PGX substrate; ●: bamboo powder. All are not significantly different at P < 0.05 by Scheffe's test.</p>

the bamboo powder substrate than in the Pro-Mix PGX substrate. In contrarst, baby blue eyes, baby's breath, brompton stock, common zinnia, cockscomb (Kurume), cypress vine, English daisy, garden balsam, pot marigold, and Roger's bronze leaf showed a higher germination rate in the Pro-Mix PGX substrate (Fig. 1). However, no statistically significant difference was observed between the two substrates in the total germination rate after 30 days for any of the species tested.

3.1.2 Pot Marigold and Pansy Cultivation

A significant difference among the substrates was observed in the main stem's length, diameter, number of leaves, and leaf SPAD of 'Orange Star' in 4 weeks after transplantation (Fig. 2). These plants had the best growth rate in the Akadama soil/peat moss substrate, and the weakest growth was recorded in the bamboo powder substrates. 'Orange Star' grown in the Akadama soil/peat moss substrate, had the highest both fresh and dry weights of shoot and root when cultivated in either Akadama soil/bamboo powder or bamboo powder substrates (Fig. 3). A similar tendency was observed for the two other pot marigold cultivars (data not shown).

A difference in both pansy height and spread due to substrate type was observed 4 weeks after transplantation (Fig. 4). These achieved the highest values 6 weeks after transplantation in the commercial potting substrate, whereas the plant spread was lowest in the bamboo powder/bamboo chip substrate. Two weeks after transplantation and later, a difference in SPAD values due to the substrate type was observed, whereas starting with 4 weeks after transplantation, the SPAD values of the pansies grown in the commercial potting substrate exceeded those of plants grown in the other substrates. The number of flowers in both the commercial potting substrate and bamboo powder/bamboo chip/bark

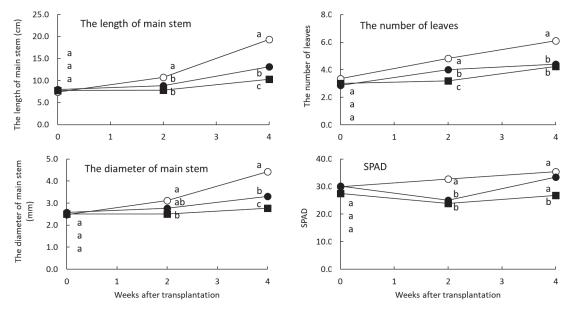


Fig. 2 Effect of different potting substrates on the growth of Pot Marigold 'Orange Star'. ○: Akadama soil/peat moss; ●: Akadama soil/bamboo powder; ■: Bamboo powder. Values followed by the same letter within a column are not significantly different at P < 0.05 by Scheffe's test.

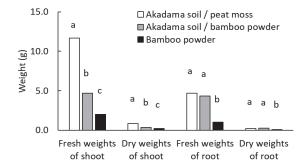


Fig. 3 Effect of different potting substrates on the weight of Pot Marigold 'Orange Star'. Values followed by the same letter within a column are not significantly different at P < 0.05 by Scheffe's test.

compost substrate were 2.5 and 2.8, respectively. The number of flowers in the bamboo powder and bamboo powder and bamboo chip substrates was 1.1 and 0.9, respectively.

From these results, it can be concluded that, although the growth and flowering of pansy in the bamboo only substrate were inferior to plants growing in the commercial potting substrate, it was possible to improve them to similar levels by mixing the bark compost with the bamboo substrate.

3.1.3 Strawberry Cultivation

The diameter of petiole, the length and width of leaf were the highest in the conventional potting substrate until week 14 after transplantation, but the difference due to substrate type had almost disappeared at week

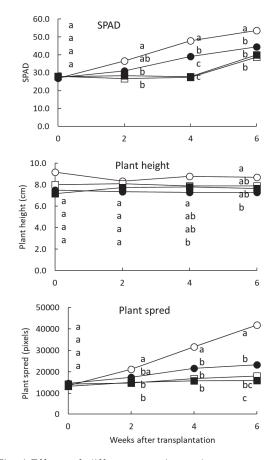


Fig. 4 Effect of different potting substrates on the growth of pansy 'Nature Blue and Yellow'. ○: Peat moss/vermiculite; □: Bamboo powder; ■: Bamboo powder/bamboo chips; ●: Bamboo powder/bamboo chips/bark compost. Values followed by the same letter within a column are not significantly different at P < 0.05 by Scheffe's test.</p>

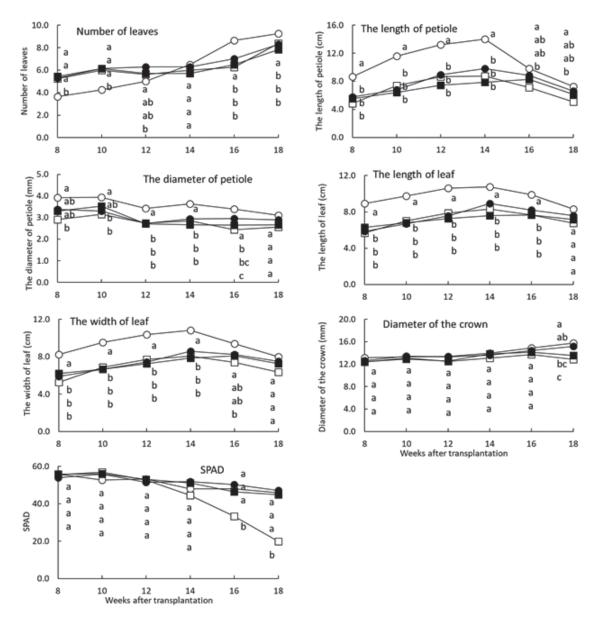


Fig. 5 Effect of different potting substrates on the growth of strawberry 'Sagahonoka'. ○: Peat moss/vermiculite; □: Bamboo powder; ■: Bamboo powder/bamboo chips; ●: Bamboo powder/bamboo chips/bark compost. Values followed by the same letter within a column <u>are</u> not significantly different at P < 0.05 by Scheffe's test.

18 after transplantation (Fig. 5). Although no difference caused by the substrate type was observed in the SPAD values until week 12 after transplantation, the SPAD of plants grown in the bamboo powder-only substrates had low values after week 16. No difference dependent on the substrate type was observed in crown diameter until week 16, but after week 18, this was the highest in plants grown in the conventional potting substrate, followed by plants grown in the bamboo powder/bamboo chip/ bark compost substrate, bamboo powder/bamboo chip substrate, and bamboo powder substrate, respectively. Leaves were most numerous in the conventional potting substrate. Fruit weight in the conventional potting substrate, in the bamboo powder substrate, in the bamboo powder/bamboo chip substrate, and in the bamboo powder/bamboo chip/compost substrate were 17.3, 20.3, 18.1 and 20.2 g, respectively. Brix of fruit in the conventional potting substrate, in the bamboo powder substrate, in the bamboo powder/ bamboo chip substrate, and in the bamboo powder/ bamboo chip/compost substrate were 8.2, 8.5, 8.9 and 7.9, respectively. These were not affected by the substrate type.

From these results, it can be concluded that, although strawberry plant growth and fruit yield in the bamboo-only substrates were inferior to those in the conventional potting substrates, the addition of bark compost to the bamboo substrates improved both strawberry plant growth and fruit yield.

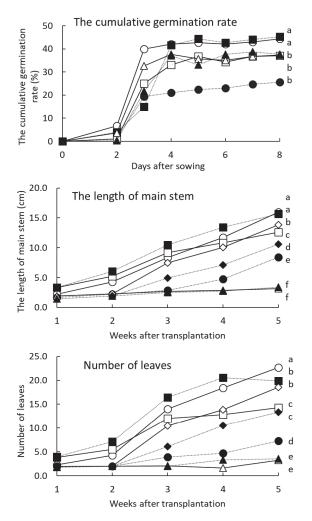


Fig. 6 Effect of different substrate recycling on the germination and growth of French marigolds 'Safari Yellow'. —○—: new; ----●----: used; —□ —: solarized; ----● =: steam-sterilized; --△—: fermented bamboo powder; ----- fermented bamboo powder; ------: unfermented bamboo powder; ------: fermented bamboo powder with fertilizer; ----- ●----: unfermented bamboo powder with fertilizer. Values followed by the same letter within a column <u>are</u> not significantly different at P < 0.05 by Scheffe's test.</p>

3.2 Substrate Recycling Using Bamboo Powder3.2.1 Plant Cultivation in Recycled Substrates

The cumulative germination rate of French marigold in new substrate was higher than in the recycled substrates (Fig. 6). Eight days after sowing, the cumulative germination rate was the highest both in the new and steam-sterilized substrates, followed by the fermented and unfermented bamboo powder substrates. A substrate with the lowest cumulative germination rate was the recycled substrate. Five weeks after transplantation, the main stem's length and number of leaves were highest in the new substrate, second lowest in the recycled substrate, and lowest in fermented and unfermented bamboo powder substrates. The main stem's length and number of leaves of plants in the fermented bamboo powder substrate with the supplied fertilizer were the highest 5 weeks after transplantation, followed by the new substrate and the steam-sterilized substrate.

3.2.2 Recycled Substrate Microbiota

Although the microbiota of both the new and solarized substrates was relatively similar, other potting substrates differed markedly among themselves in the microbiota composition (Table 1). The fermented bamboo powder substrate contained a high proportion of microbes, including Actinobacteria and Bacteroidetes belonging to the following genera: *Sphingobacterium, Anaerobacillus,* and *Ochrobactrum.* In the steam-sterilized substrate, Firmicutes belonging to the genus *Bacillus* and unidentifiable DNA fragments accounted for approximately 36.2% and 44.3% of the total microbes, respectively.

4. Discussion

Both germination and growth of the flowering plants potted in bamboo substrates were found generally inferior to those of plants grown in the conventional potting substrate. However, in the growth of pansies improved by the addition of compost to a bamboo substrate. Furthermore, French marigolds show better growth when a fertilizer and bamboo powder are simultaneously provided as an amendment to a recycled potting substrate. Gruda et al. (2000) reported that wood fiber substrates provide microorganisms with the mineral nitrogen they require to build up their protein components. However, nitrogen would not be readily available for plants growing in wood fiber substrates. Generally, nitrogen shortage in the soil arises as the nitrogen is absorbed during the decomposition of the organic matter. By investigating the effect of using bamboo compost during cultivation of soybean (Nakagawa et al., 2009; Yamakawa et al., 2009a; Yamakawa et al., 2009b), it was reported that the mulch of bamboo powder increased the seed yield more than the mixing of bamboo powder in soil. The mixing of bamboo powder in soil decreases the seed yield less than nomixing. Thus, it was though that the initial growth after mixing bamboo powder in soil depressed and the

	New substrate (%)	Used substrate (%)	S olarized substrate (%)	Steam-sterilized substrate (%)	Fermented bamboo powder substrate (%)	Unfermented bamboo powder substrate (%)
unidentifiable DNA fragments	1.6	0.4	1.0	44.3	1.4	3.9
Crenarchaeota	0.5	0.2	0.4	0.0	0.0	0.6
Acidobacteria	8.4	0.1	6.9	0.0	0.0	4.0
Actinobacteria	18.4	7.9	20.3	0.6	35.9	11.7
Armatimonadetes	0.4	0.0	0.4	0.0	0.0	0.1
Bacteroidetes	5.0	5.2	3.0	0.1	11.6	2.0
Chlamydiae	0.7	0.1	0.7	0.0	0.0	0.5
Chlorobi	0.2	0.0	0.3	0.0	0.0	0.1
Chloroflexi	5.3	0.3	5.7	0.0	0.1	2.9
Cyanobacteria	0.3	0.3	0.3	0.0	0.0	0.1
Firmicutes	9.0	63.4	14.8	36.2	9.9	45.3
Gemmatimonadietes	1.5	0.2	1.4	0.0	0.0	0.7
Nitrospirae	0.3	0.0	0.3	0.0	0.0	0.3
Planctomycetes	4.0	0.4	4.0	0.0	0.0	2.3
Proteob acteria	41.1	19.5	38.6	18.8	41.0	24.2
Verrucomicrobia	1.2	1.9	0.8	0.0	0.0	0.5

Table 1 Microbiota composition in 6 substrate

growth rate was equally recovered to that for surface application after flowering. However, it did not reflect on the actual seed production. They pointed out that this observation may be due to nitrogen starvation. This phenomenon occurs when the nitrogen in the soil is absorbed, as the bamboo compost contains few amounts of fertilizer. In this study, by adding fertilizer to the bamboo substrates, microbes decomposing bamboo and the horticultural plants grown in it competed for nitrogen. In such a scenario, there was a possibility that the horticultural plants grown in bamboo substrates are starved of nitrogen. If the nitrogen starvation in bamboo substrates is established, the growth suppression in bamboo substrates is solved, and bamboo substrates could find a commercial application.

When using the main stem length and number of leaves of French marigolds as indices, the recycled substrate that was the closest to the unused commercial substrate, in terms of its performance, was the substrate that had been steam-sterilized for two hours at 100°C. However, amplicon analysis revealed that this type of substrate contains several unidentifiable DNA fragments. This is because steam sterilization of a substrate using a steam boiler kills many soil-dwelling microbes, meaning that there is a high possibility of it disturbing the balance of substrate microbiota. Consequently, certain pathogenic microbe strains can multiply in a substrate. Therefore, steam sterilization can hardly be considered the best way to revive the recycling substrates. Conversely, recycled Pro-Mix PGX substrate with 30% fermented bamboo powder was found to contain many Actinobacteria. Actinobacteria prefer cold places

(leaving under leaves) and weather and are said to be involved in compost fermentation. Moreover, if chitin amount is increased in the soil, then Actinobacteria increase, and their activation is considered to suppress the activity of pathogens such as filamentous fungi. If fertilizer is added to fermented bamboo powder substrates, both the main stem length and number of leaves of French marigolds increase at a rate that is inferior only to the new and steam-sterilized substrates. Although much attention has to be paid to finding the best possible ways for fertilizer application, fermented bamboo powder substrates are promising candidates for commercial recycling of used potting substrates.

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