

# Effects of Applying Bagasse-Based Spent Mushroom Substrate on Early Growth of Sugarcane and Reduction of Environmental Impact

Takashi KANDA<sup>1\*</sup>, Yoshifumi TERAJIMA<sup>1</sup>, Kenji TAMURA<sup>2</sup>, Hiroko NAKATSUKA<sup>3</sup> and Satoshi NAKAMURA<sup>4</sup>

<sup>1</sup> Tropical Agriculture Research Front, Japan International Research Center for Agricultural Sciences, Ishigaki, Japan

<sup>2</sup> Faculty of Life and Environmental Sciences, University of Tsukuba, Tsukuba, Japan

<sup>3</sup> Faculty of Agriculture, Tokyo University of Agriculture, Atsugi, Japan

<sup>4</sup> Crop, Livestock and Environment Division, Japan International Research Center for Agricultural Sciences, Tsukuba, Japan

## Abstract

Applying spent mushroom substrate (SMS) in sugarcane farming can be an effective use of organic resources, promoting growth while improving soil fertility. However, most SMS is disposed of as waste, and application methods have not been adequately studied. Therefore, we verified the effects of SMS made from bagasse (SMS-B) on sugarcane and its potential to reduce environmental impacts, such as chemical fertilizer use and climate change, comparing its application with that of chemical fertilizers (CF), bagasse, and SMS from sawdust (SMS-S). The SMS-B application increased dry matter weight by producing more tillers than CF. Additionally, SMS-B improved many soil chemical properties compared with CF. SMS-B promotes the early growth of sugarcane because it contains more nutrients and has a lower C:N ratio than bagasse or SMS-S, likely due to differences in the materials used for mushroom cultivation, as SMS-B was made from composted bagasse. With a 30% CF reduction, SMS-B achieved a higher dry matter weight than the recommended CF application. Compared to the other treatments, SMS-B resulted in higher total soil carbon content relative to the amount of applied carbon. These results indicate that the application of SMS-B can promote the early growth of sugarcane and effectively reduce environmental impacts via reducing chemical fertilizer use and increasing total soil carbon content.

**Discipline:** Agricultural Environment

**Additional key words:** chemical fertilizer reduction, resource recycling, soil chemical properties, tillers, tropical islands

## Introduction

With the growing trend toward healthy eating, mushrooms have become popular additions to daily meals due to their high nutritional content (Umor et al. 2021, Valverde et al. 2015). Mushroom production has grown worldwide at a compound annual growth rate of 8.26% since 2000, increasing more than fivefold (Bijla & Sharma 2023). Generally, mushrooms are cultivated on agricultural waste, such as rice and wheat waste, which is a reliable source of nutrition, as the substrate composition significantly influences mushroom yield (Jayaraman

et al. 2024). However, mushroom cultivation generates large amounts of spent mushroom substrate (SMS), with 1 kg of fresh mushrooms producing approximately 5 kg of SMS (Leong et al. 2022, Umor et al. 2021). Around 104 million tons of SMS are expected to be generated worldwide per year by 2026 (Atallah et al. 2021). This waste is typically disposed of in landfill sites or incinerated, resulting in significant environmental pollution (Jiang et al. 2017, Williams et al. 2001). Therefore, attempts have been made to use SMS as an organic fertilizer and source of biogas (Jayaraman et al. 2024, Pérez-Chávez et al. 2019).

---

\*Corresponding author: [kandat0322@jircas.go.jp](mailto:kandat0322@jircas.go.jp)

Received 19 March 2025; accepted 28 July 2025; J-STAGE Advanced Epub 28 January 2026.

<https://doi.org/10.6090/jarq.24J32>

The use of SMS from the cultivation of different mushrooms (*Agaricus bisporus*, *Agaricus blazei*, Enoki, and *Auricularia auricula*) as organic fertilizer on farmland has been shown to be effective for crop production and the soil environment, particularly for soil chemical and biological properties (Carpio et al. 2023, Chen et al. 2022, Li et al. 2020, Yoshimoto et al. 2016). Furthermore, studies on SMS from *Pleurotus ostreatus* cultivation have found that it not only affects the soil nutrient content of the soil surface layer (Oda et al. 2014) but also improves the soil structure of the underlying layer (Nakatsuka et al. 2016). This shows it is effective for sustainable agriculture. In addition to increasing crop yield and improving the soil environment, the application of SMS to agricultural land reduces environmental impacts, such as heavy metal contamination, and mitigates climate change (Chen et al. 2022, Li et al. 2020).

The sugarcane (*Saccharum* spp. Hybrids) industry is significant on tropical islands. Bagasse is the residue produced after sugarcane juice extraction, and its main components are cellulose, pentosan, lignin, and ash. Most bagasse is used as fuel for boilers in sugar mills; other uses include the production of pulp and paper, hardboards, feed and forage yeast, furfural, and fertilizers (Miyazato 1986). Bagasse is also used as a substrate for mushroom cultivation, having proven effective in Japan (Yamauchi et al. 2015). Additionally, it has been utilized as a substrate for cultivating *Auricularia nigricans* on subtropical islands (Hira 2020). Although some SMS made from bagasse (SMS-B) is returned to the fields, much remains unused. SMS-B has the potential to be a valuable organic resource for enhancing sugarcane production. However, few studies have demonstrated the effectiveness of SMS-B in this regard.

In Japan's southwestern islands, the country's main sugarcane production area, missing plants caused by waterlogging or improper fertilization management and typhoon-induced breakage contribute to low sugarcane yields (Sugimoto et al. 2003). Increasing plant size during early growth in spring planting and ratooning is vital to minimize damage caused by typhoons (Miyagi et al. 2002). Applying organic materials can promote early growth, with the introduction of green manure before planting increasing the number of stems (Tarora et al. 2005). Furthermore, organic matter application can help stabilize yields by lowering missing plant rates in ratoon crops (Yokoi et al. 2005).

Localized heavy rainfall caused by climate change may lead to the discharge of large amounts of sediment and soil nutrients from sugarcane fields, particularly during the early growth phase (Thomaz et al. 2022). The resulting eutrophication of coral reefs owing to this

excessive nutrient runoff is a concern on tropical islands (Nakanishi 2017). Load reduction and carbon sequestration in sugarcane fields are essential for the environmental conservation of tropical islands. In addition, the MIDORI Strategy for Sustainable Food Systems prepared by the Ministry of Agriculture, Forestry and Fisheries in Japan set the goal of reducing chemical fertilizer (CF) use 30% by 2050, calling for the development of technology to reduce it while maintaining productivity. Returning SMS-B to farmland is a promising aid to sustainable sugarcane production and also for resource recycling and reducing environmental impacts on tropical islands.

Therefore, this study conducted a pot experiment in Kunigami marge soil, a highly weathered and widely distributed soil in Okinawa Prefecture, to clarify (1) the effect of SMS-B application on the early growth of sugarcane and soil chemical properties compared with SMS application of sawdust and CFs, and (2) the possibility of reduced CF use and soil carbon content by applying SMS-B.

## Materials and methods

### 1. Pot experiment and measurement of soil water content

The pot experiment was conducted from 18 May to 18 October 2022 (five months) in a greenhouse at the Tropical Agriculture Research Front, JIRCAS, in Ishigaki, Okinawa, Japan. The mean temperature and humidity in the greenhouse during the experiment were 28.7°C and 67.6%, respectively.

The pots used were 1/2000a Wagener pots (25.2 cm in diameter and 30 cm in height) with bottom holes. The study used approximately 15 kg pot<sup>-1</sup> of Kunigami marge soil (mainly classified as Alisols, Acrisols, or Cambisols according to the World Reference Base for Soil Resources), which is widely distributed in the southwestern islands of Japan. The soil was air-dried and passed through a 6 mm sieve prior to the experiment. Each pot was filled to a soil depth of 24.5 cm. The initial chemical properties of the soil are listed in Table 1. CFs and organic materials were mixed with the soil before filling them. After that, water was supplied up to 60% of the field water capacity.

The Ni27 Japanese sugarcane cultivar, the most widely one grown in Okinawa Prefecture, was used as the test crop. Three single-bud setts were planted in each pot at a depth of 10 cm. Two weeks after planting, seedlings were thinned, leaving only the tallest plant per pot.

The study consisted of five treatments: (T1) chemical fertilizer (CF) as a control, (T2) CF and bagasse, (T3) CF

**Table 1. Initial soil properties of soil used for the pot experiment**

Soil property	Unit	Amount	
Clay	%	33.0	
Silt	%	17.6	
Sand	%	49.4	
pH(H <sub>2</sub> O)		5.30	
EC	mS m <sup>-1</sup>	6.57	
TN	%	0.07	
TC	%	0.75	
C:N		10.63	
CEC	cmolc kg <sup>-1</sup>	16.06	
Ex-Ca		4.36	
Ex-Mg		1.08	
Ex-K		0.37	
Ex-Na		0.20	
Ava-P <sub>2</sub> O <sub>5</sub>	Bray1	mgP <sub>2</sub> O <sub>5</sub> kg <sup>-1</sup>	4.36
Ava-P <sub>2</sub> O <sub>5</sub>	Bray2	mgP <sub>2</sub> O <sub>5</sub> kg <sup>-1</sup>	15.37
Ava-SiO <sub>2</sub>		mgSiO <sub>2</sub> kg <sup>-1</sup>	13.95
Ava-N		mgN kg <sup>-1</sup>	26.55

and SMS from sawdust (SMS-S), (T4) CF and SMS-B, and (T5) 70% CF and SMS-B.

The bagasse used for the pot experiment was provided by Ishigakijima Sugar Co. SMS-S was applied to shiitake mushroom (*Lentinula edodes*) cultivation; the culture material consisted of uncomposted sawdust, corn, and rice bran. SMS-B was used for cultivating wood ear mushroom (*Auricularia auricula-judae*), and the culture material was bagasse that had been composted for one year. Then, SMS-B was stored outdoors under a roof for four months before the experiment. These organic materials were oven-dried at 70°C and pulverized in a mixer for use in powder form. Sawdust is commonly used as a mushroom substrate for shiitake mushrooms (Forest Resources Research Center, Department of Agriculture, Forestry and Fisheries, Okinawa Prefecture 2022). This study did not consider differences in mushroom varieties but did compare different SMSs (SMS-B and SMS-S). The components of each type of organic material are listed in Table 2.

The treatments with these organic materials (T2–T5) were divided into two groups with different amounts of applied organic material (30 t ha<sup>-1</sup> and 60 t ha<sup>-1</sup> dry matter weight). Each treatment was conducted with five replicates (45 pots in total).

CF was applied according to the sugarcane cultivation guidelines of Okinawa Prefecture (Department of Agriculture, Forestry and Fisheries in Okinawa

**Table 2. Elemental composition of organic materials used for the pot experiment**

		Organic material		
		Bagasse	SMS-S	SMS-B
Water	%	9.51	9.69	11.79
Ash	%	4.56	5.78	28.72
pH(H <sub>2</sub> O)		7.0	4.2	7.4
T-N	%	0.24	0.90	1.50
T-C	%	45.62	40.94	30.69
C:N		189.1	45.6	20.5
T-P <sub>2</sub> O <sub>5</sub>	%	0.09	1.40	2.60
T-K <sub>2</sub> O	%	0.20	0.49	1.36
T-CaO	%	0.63	2.04	6.85
T-MgO	%	0.07	0.56	1.04
T-SiO <sub>2</sub>	%	2.60	0.31	10.57

Water content is shown as % of actual product. Others are shown as % of dry product.

Prefecture 2014). Before planting, 0.643 g pot<sup>-1</sup> of urea (60 kg N ha<sup>-1</sup>) was applied as N, 0.322 g pot<sup>-1</sup> of triple superphosphate as P<sub>2</sub>O<sub>5</sub> (30 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>), and 0.237 g pot<sup>-1</sup> of potassium chloride as K<sub>2</sub>O (30 kg K<sub>2</sub>O ha<sup>-1</sup>). The same amount of each fertilizer was applied two months after the start of the experiment. For the 70% CF treatment (T5), 70% of each chemical fertilizer was applied simultaneously with the other treatments (Table 3). During the experimental period, the pots were watered two or three times a week until the middle stage of growth, and then daily to prevent the soil from drying out. From the middle of the experiment, the soil water conditions within the pots differed for each treatment, so the soil water content was measured at the surface using a soil moisture sensor (TEROS 12, METER). Measurements were taken once a month, with one pot from each treatment selected for the days after seeding (DAS) 46 and DAS77 and all pots for DAS120 and DAS141.

## 2. Sugarcane growth survey and analysis

During the experiment, monthly surveys were conducted to measure sugarcane growth, including culm length and the number of sugarcane tillers. After the experiment, the aboveground parts of the plants were harvested and divided into millable stalks and other parts, such as leaves and cane tops. The lengths, diameters, number of nodes, and Brix values of the millable stalks were measured. Using digital calipers, the stalk diameter was measured at the minor axis of the node located nearest the middle of the stalk. The Brix value was investigated using a hand-held refractometer (MASTER-Pa, Atago Co., Ltd., Tokyo, Japan) using cane

**Table 3. Amounts of each component from chemical fertilizers and organic materials added to each treatment**

Treatment	Amount (t ha <sup>-1</sup> )	Chemical fertilizer*			Organic materials			Total		
		N	P <sub>2</sub> O <sub>5</sub> (kg ha <sup>-1</sup> )	K <sub>2</sub> O	N	P <sub>2</sub> O <sub>5</sub> (kg ha <sup>-1</sup> )	K <sub>2</sub> O	N	P <sub>2</sub> O <sub>5</sub> (kg ha <sup>-1</sup> )	K <sub>2</sub> O
CF	0	120	60	60	0	0	0	120	60	60
Bagasse		120	60	60	65	24	54	185	84	114
SMS-S	30	120	60	60	243	378	132	363	438	192
SMS-B		120	60	60	397	687	360	517	747	420
SMS-B (CF70%)		84	42	42	397	687	360	481	729	402
Bagasse		120	60	60	131	48	108	251	108	168
SMS-S	60	120	60	60	487	756	264	607	816	324
SMS-B		120	60	60	793	1,374	720	913	1,434	780
SMS-B (CF70%)		84	42	42	793	1,374	720	877	1,416	762

CF: chemical fertilizer, SMS-S: SMS from sawdust, SMS-B: SMS from bagasse

The amount of each component of chemical fertilizer is shown as the total amounts of first and second doses.

juice collected from the middle internode. Finally, the millable stalk and other parts were crushed, dried at 70°C, and weighed as dry matter. The total dry matter weight was calculated as the sum of the dry matter weight of the millable stalks and other parts.

### 3. Soil sampling and soil analysis

The soil used for chemical analysis was mixed after sugarcane harvesting, and a portion was collected and air-dried. The soil was then passed through a 2 mm sieve and analyzed.

The pH (H<sub>2</sub>O) and EC were measured using the glass electrode method (LAQUA F-72 and ES-51; HORIBA Ltd., Kyoto, Japan) at a soil solution ratio of 1:5. The total nitrogen (TN) and carbon (TC) contents were measured using the dry combustion method using an NC analyzer (NC 220F; Sumika Chemical Analysis Service, Tokyo, Japan). Exchangeable cations (Ca, Mg, K, and Na) were determined by ICP (ICPE-9820; Shimadzu Corp., Osaka, Japan) using 1 M ammonium acetate as the extractant. After the exchangeable cations were extracted, the soil residues were washed with 80% methanol and then extracted with 10% KCl. The NH<sub>4</sub>-N content of the extracts was measured using an autoanalyzer (Auto Analyzer III, BL-TEC K. K., Tokyo, Japan) to calculate the cation exchangeable capacity (CEC). For available nitrogen (Ava-N), soils were incubated at 30°C for four weeks and extracted using 2 M KCl. After the Kjeldahl distillation of the extract, titrations were performed to determine the amount of available nitrogen (Committee on Soil Environmental Analysis Methods 1997). The Bray1 and Bray2 methods were used to measure available phosphate (Ava-P<sub>2</sub>O<sub>5</sub>). Each extract, prepared with a different solvent, was analyzed using a spectrophotometer

(UV-1800; Shimadzu Corp., Osaka, Japan; Committee on Soil Nutrient Determination Methods, 1970). Available silicate (Ava-SiO<sub>2</sub>) was extracted according to the phosphate buffer extraction method (Soil Association of Japan 2001) and analyzed using a spectrophotometer (UV-1800; Shimadzu Corp., Osaka, Japan).

### 4. Statistical analyses

Differences between treatments in sugarcane growth, dry matter weight, millable stalk characteristics, and soil chemical properties were determined using Dunnett's test with the CF treatment used as the control. Statistical analysis was performed using JMP 17.2.0 (JMP Statistical Discovery LLC.).

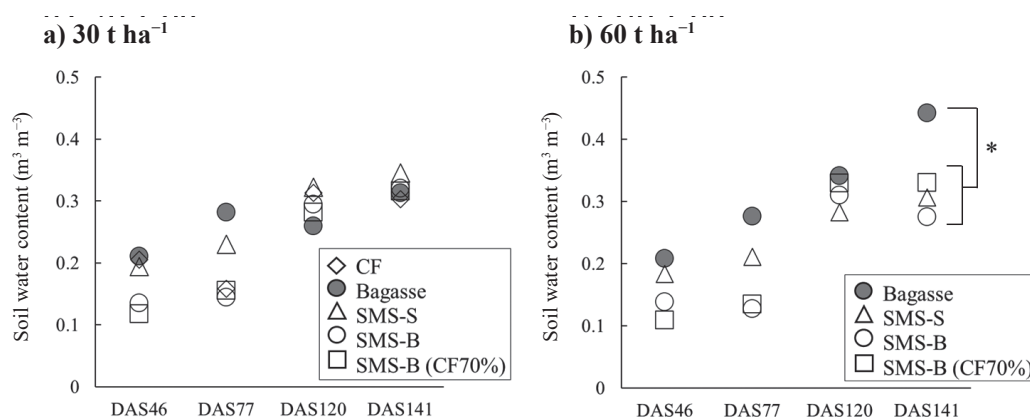
## Results

### 1. Soil water content

Figure 1 shows the soil water content for each treatment. During the experiment (DAS46 and DAS77), the soil water content was higher in the bagasse and SMS-S treatments than in the others, regardless of the amount applied. By the end of the experiment (DAS120 and DAS141), no significant overall differences were observed; however, the soil water content of the bagasse treatment applied at 60 t ha<sup>-1</sup> in DAS141 was significantly higher ( $P < 0.05$ ). In the bagasse treatment, waterlogging of the soil surface was observed after irrigation.

### 2. Sugarcane growth during the experiment

Table 4 shows the changes in culm length and the number of sugarcane tillers for each treatment. The change in culm length tended to be lower for bagasse than for other treatments; however, no significant



**Fig. 1. Soil water content in each treatment during the pot experiment**

CF: chemical fertilizer, SMS-S: SMS from sawdust, SMS-B: SMS from bagasse

Asterisks indicate significant differences ( $p < 0.05$ ).

DAS46 and DAS77 data are representative for each treatment ( $n=1$ ), and those of DAS120 and

DAS141 are averages of five pots for each treatment.

**Table 4. Changes in culm length and the number of tillers during the pot experiment**

Treatment	Amount (t ha <sup>-1</sup> )	Culm length (cm)					Number of tillers				
		DAS29	DAS65	DAS90	DAS120	DAS160	DAS29	DAS65	DAS90	DAS120	DAS160
CF	0	21.8	76.9	119.4	185.7	236.4	0	0	0.3	1.2	1.4
Bagasse		21.1	41.4**	80.9**	130.6**	170.0**	0	0	0	0	0
SMS-S	30	19.2	62.5	114.6	178.2	228.8	0	0	0	0	0
SMS-B		23.1	80.8	118.5	188.1	238.8	0	2.5**	2.6**	2.8	2.8
SMS-B (CF70%)		19.3	83.3	121.4	190.5	240.6	0	1.8**	1.8*	1.8	1.8
Bagasse		12.1**	17.3**	39.5**	84.1**	122.6**	0	0	0	0	0
SMS-S	60	16.6	60.9	117.7	183.3	240.4	0	0	0.2	0.2	0.8
SMS-B		21.0	73.4	108.7	186.3	241.2	0	3.0**	3.4**	3.0	3.2
SMS-B (CF70%)		23.7	76.1	112.3	186.3	239.6	0	3.2**	3.2**	3.0	3.2

CF: chemical fertilizer, SMS-S: SMS from sawdust, SMS-B: SMS from bagasse

DAS indicates day after seeding.

Asterisks indicate significant differences (\*\*  $p < 0.01$ , \*  $p < 0.05$ ).

Statistical analysis was done by Dunnett test with CF as control.

differences were observed among the other treatments after DAS90. In contrast, the number of tillers increased after two months of planting (DAS65) in the SMS-B and SMS-B (CF70%) treatments. It was higher at 60 t ha<sup>-1</sup> than at 30 t ha<sup>-1</sup> when compared with the amount applied. CF and SMS-S at 60 t ha<sup>-1</sup> also increased the number of tillers from three months after planting (DAS90). However, no tillers were observed in the bagasse or SMS-S with the 30 t ha<sup>-1</sup> treatments.

### 3. Dry matter weight of sugarcane and millable stalk characteristics

The total dry matter and millable stalk weights for each treatment are shown in Table 5. These weights were highest in the SMS-B and SMS-B (CF70%) treatments,

followed by SMS-S, CF, and bagasse. A higher amount of applied organic material correlated with higher weights. However, dry weight was lower when treated with 60 t ha<sup>-1</sup> of bagasse.

The agronomic traits of the millable stalks (stalk length, diameter, internode number, and Brix values; Table 5) were lower in the bagasse treatment than in CF. Comparing the amount of applied organic materials, the 60 t ha<sup>-1</sup> application resulted in lower agronomic traits of the millable stalks than the 30 t ha<sup>-1</sup> application in the bagasse treatment, similar to the dry matter weight results. No clear differences were observed among the other treatments.

**Table 5. Agronomic traits of millable stalk in each treatment**

Treatment	Amount (t ha <sup>-1</sup> )	TDMW (g pot <sup>-1</sup> )	MSW (g pot <sup>-1</sup> )	Stalk length (cm)	Stalk diameter (mm)	Internode number (no. stalk <sup>-1</sup> )	Brix value (° Brix)
CF	0	296.7 ± 19.7	184.1 ± 7.9	165.8 ± 8.2	22.4 ± 0.7	12.6 ± 0.9	21.3 ± 0.8
Bagasse		128.2 ± 8.3**	67.4 ± 6.5**	106.4 ± 2.6**	17.2 ± 0.5**	9.0 ± 0.7**	19.7 ± 1.0
SMS-S	30	267.1 ± 30.6	164.1 ± 31.8	164 ± 12.0	20.7 ± 0.5	12.4 ± 0.5	20.4 ± 2.2
SMS-B		390.3 ± 13.6**	216.3 ± 21.5	166.6 ± 10.4	22.7 ± 1.2	13.8 ± 0.8	17.6 ± 2.2*
SMS-B (CF70%)		361.8 ± 19.7**	210.9 ± 18.1	169.4 ± 11.3	21.4 ± 1.9	13.2 ± 0.4	19.2 ± 2.3
Bagasse		66.4 ± 9.3**	27.5 ± 4.8**	57.8 ± 7.8**	14.1 ± 0.4**	5.0 ± 1.0**	16.5 ± 1.5**
SMS-S	60	306.5 ± 24.1	189.0 ± 22.5	170 ± 10.9	21.8 ± 0.9	13.0 ± 0.0	20.8 ± 1.1
SMS-B		453.4 ± 29.1**	250.3 ± 19.1**	164.8 ± 9.7	22.8 ± 1.4	14.0 ± 0.7*	18.0 ± 3.2
SMS-B (CF70%)		438.1 ± 32.4**	228.9 ± 18.6**	162.2 ± 10.3	21.8 ± 1.3	13.6 ± 0.5	18.4 ± 1.7

CF: chemical fertilizer, SMS-S: SMS from sawdust, SMS-B: SMS from bagasse

TDMW and MSW indicate total dry matter weight and millable stalk weight.

Asterisks indicate significant differences (\*\*  $p < 0.01$ , \*  $p < 0.05$ ).

Statistical analysis done by Dunnett test with CF as control.

**Table 6. Soil chemical properties in each treatment after the pot experiment**

Treatment	Amount (t ha <sup>-1</sup> )	pH(H <sub>2</sub> O)	EC	TN	TC	C:N	CEC	Ex-Ca	Ex-Mg	Ex-K	Ex-Na	Ava-P <sub>2</sub> O <sub>5</sub>		Ava-SiO <sub>2</sub>	Ava-N
												Bray1	Bray2		
			(mS m <sup>-1</sup> )	(%)	(%)			(emolc kg <sup>-1</sup> )			(mgP <sub>2</sub> O <sub>5</sub> kg <sup>-1</sup> )	(mgSiO <sub>2</sub> kg <sup>-1</sup> )	(mgN kg <sup>-1</sup> )		
CF	0	5.91	6.47	0.09	0.82	9.45	13.57	6.23	1.09	0.14	0.50	3.39	10.73	130.7	7.53
Bagasse		5.88	6.28	0.10**	1.09**	10.56	12.94	5.95	1.10	0.21**	0.41	2.38	9.74	173.1**	n.d.
SMS-S	30	6.16**	6.84	0.11**	1.00*	8.83	12.87	6.46	1.29**	0.17*	0.50	4.80	16.22	141.3	15.65**
SMS-B		6.45**	6.37	0.11**	1.27**	11.40**	13.82	8.26**	1.35**	0.13	0.57	10.57**	24.58**	126.0	11.29
SMS-B (CF70%)		6.54**	6.57	0.12**	1.25**	10.50	15.88**	8.33**	1.36**	0.14	0.54	10.07**	28.19**	123.7	11.44
Bagasse		5.61**	6.57	0.11**	1.37**	12.20**	12.13	5.05**	0.89**	0.31**	0.35**	1.26	8.53	206.9**	n.d.
SMS-S	60	6.23**	7.30*	0.12**	1.26**	10.74*	14.25	7.01**	1.45**	0.21**	0.53	7.83**	21.50**	128.8	12.18
SMS-B		6.69**	7.66**	0.13**	1.62**	12.23**	14.59	10.00**	1.51**	0.16	0.66**	16.10**	37.86**	141.8	17.95**
SMS-B (CF70%)		6.64**	7.77**	0.14**	1.68**	11.94**	16.42**	10.23**	1.63**	0.18**	0.54	15.53**	38.18**	138.3	18.07**

CF: chemical fertilizer, SMS-S: SMS from sawdust, SMS-B: SMS from bagasse

Different letters indicate significant differences ( $p < 0.05$ ).

n.d. indicates not detected.

Asterisks indicate significant differences (\*\*  $p < 0.01$ , \*  $p < 0.05$ ).

Statistical analysis was done by Dunnett test with CF as control.

#### 4. Effects of organic material application on soil chemical properties

The soil chemical properties for each treatment are shown in Table 6. SMS-B treatment at 60 t ha<sup>-1</sup> showed higher values for many soil chemical properties. In particular, pH(H<sub>2</sub>O), EC, TN, TC, C:N ratio, exchangeable Ca, Mg, Na, available phosphate, and available nitrogen were significantly higher than those of CF. The CF and bagasse treatments showed lower values for many soil chemical properties, whereas SMS-S values were intermediate. Although many soil chemical properties of the bagasse treatment were lower, including TN, TC, and the C:N ratio, the exchangeable K and available silicate

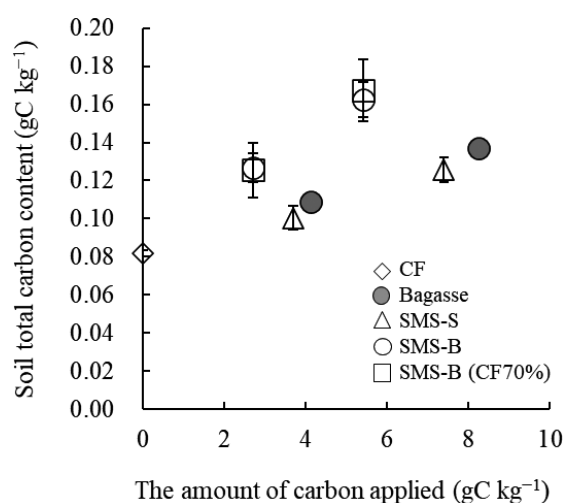
were higher than those of CF.

Figure 2 shows the relationship between the amount of applied carbon and the total soil carbon content for each treatment. Compared to the others, the SMS-B treatment resulted in higher soil total carbon content relative to the amount of carbon applied.

## Discussion

### 1. Effects of SMS-B application on the early growth of sugarcane

The dry matter weights of sugarcane (total dry matter and millable stalk weights) were higher with the



**Fig. 2. Relationship between the amount of carbon applied and soil total carbon content in each treatment**

CF: chemical fertilizer, SMS-S: SMS from sawdust, SMS-B: SMS from bagasse

application of SMS-B than CF, regardless of the amount applied (Table 5). This result was consistent with those of previous studies; Yoshimoto et al. (2016) applied 20 t ha<sup>-1</sup> of SMS from sugarcane shoots after *Himematsutake* (*Agaricus blazei* Murill) mushroom production to paddy soil, resulting in a 14.8% increased aboveground weight of forage rice cultivars compared to the usual CF application. Gobbi et al. (2018) showed that the application of SMS made from straw + poultry manure, straw + horse manure + poultry manure, and horse manure + poultry manure as a substitute for nitrogen fertilizers for lettuce and leeks resulted in yields comparable to those obtained when using CFs. As shown in Table 2, SMS-B contained more nitrogen, phosphate, and potassium as fertilizer components than bagasse and SMS-S.

The increase in dry matter weight (total dry matter and millable stalk weight) with SMS-B application was attributed to an increased number of tillers (Table 4). Regarding the factors influencing the number of tillers, Vuyyuru et al. (2019) compared the differences in tiller numbers and sugarcane yield between nitrogen fertilizer application levels and timing in Histosols in Florida, USA. They found that more nitrogen resulted in a greater number of tillers in both plant and ratoon crops. Yoshida et al. (2017) also showed that applying molasses increased the yields of plant and ratoon crops, as well as the number of tillers, and concluded that an increase in the amount of available nitrogen in the soil promoted an increase in the number of tillers. Thus, the nitrogen supply from SMS-B and the increased available nitrogen in the soil due to the addition of SMS-B may have contributed to the higher

number of tillers, resulting in the higher dry matter weights of sugarcane observed in this study.

## 2. Effect of different organic material applications on the early growth of sugarcane

Bagasse application resulted in a lower dry matter weight of sugarcane than using CF, despite the nutrient supply from bagasse (Tables 2, 5). During the experiment, the soil water content in the bagasse treatment was higher than that in the other treatments, particularly at the end of the experiment (Fig. 1). The dry matter weights of sugarcane were significantly lower with 60 t ha<sup>-1</sup> application (Table 5), indicating that poor drainage led to poor germination and a reduced number of tillers (Shinzato et al. 2013). We believe this is one of the factors contributing to poor sugarcane growth. In addition, bagasse was characterized by a high C:N ratio (189.1, Table 2). When an organic material with a high C:N ratio is applied, soil microorganisms utilize its inorganic nitrogen for growth, decreasing the amount of inorganic nitrogen in the soil (Kyuma 1997). This is evident from the fact that the bagasse application resulted in no available nitrogen (Table 6). In previous studies, applying bagasse after composting (with a C:N ratio of 20) has been shown to increase crop yield (Inoue & Hashiguchi 2011, Kuba et al. 1989). The SMS-B used in this study was made from bagasse that had been composted for approximately one year and then stored for an additional four months before use. For this reason, SMS-B had a lower C:N ratio, making it an effective organic material for the early growth of sugarcane.

Compared with the CF treatment, neither the 30 t ha<sup>-1</sup> nor the 60 t ha<sup>-1</sup> application of SMS-S had a positive effect on the dry matter weight of sugarcane, despite the nutrient supply from SMS-S (Tables 2, 5). In addition, as with SMS-B, the amounts of available nitrogen and phosphate in the soil increased with the amount added (Table 6). However, unlike SMS-B, the addition of SMS-S did not increase the number of tillers (Table 4). Furthermore, the amount of available nitrogen in the 30 t ha<sup>-1</sup> SMS-S treatment was higher than in the 60 t ha<sup>-1</sup> SMS-S treatment and the 30 t ha<sup>-1</sup> SMS-B treatment (Table 6). The absence of a positive effect of the SMS-S application on sugarcane growth was because the C:N ratio of SMS-S (C:N ratio: 45.6) was higher than that of SMS-B (C:N ratio: 20.5). Kumada (1977) classified nitrogen mineralization from organic materials into three groups: Group 1 with C:N ratios of 5.6-11.3 (the mineralization of organic nitrogen occurs immediately after incubation and then declines); Group 2 with C:N ratios of 15.3-20.6 (the immobilization of soil-derived inorganic nitrogen occurs initially, followed by organic

nitrogen mineralization); and Group 3 with C:N ratios of 37.1-64.4 (immobilization occurs over 30 days, followed by a slow mineralization). The C:N ratio of SMS-S was 45.6, falling within Group 3, indicating that nitrogen immobilization occurred in the early stages of the experiment, similar to that observed in bagasse. Since nitrogen fertilization or available nitrogen in soil contributed to the increase in the number of tillers (Vuyyuru et al. 2019, Yoshida et al. 2017), the lack of observed tillers when SMS-S was applied can be attributed to the low amount of inorganic nitrogen from organic materials in the early growth stage. In addition, the amount of available nitrogen in the 60 t ha<sup>-1</sup> SMS-S application was lower than in the 30 t ha<sup>-1</sup> application, which could be attributed to the longer immobilization of nitrogen following the 60 t ha<sup>-1</sup> application.

### 3. Potential for reducing chemical fertilizer (CF) use and carbon sequestration by application of SMS-B

The addition of SMS-B with a 30% reduction in chemical fertilizer (SMS-B (70%)) increased the dry matter weight of sugarcane in both the 30 t ha<sup>-1</sup> and 60 t ha<sup>-1</sup> treatments over the CF treatment (Table 5). Li et al. (2020) investigated the effects of a ten-year application of SMS from *Agaricus bisporus* in rice cultivation and demonstrated that applying SMS can reduce the use of CFs and improve soil quality. The results of this study are consistent with those of previous studies, indicating that SMS-B can be used as an alternative to CFs.

In addition, based on the nutrient content in SMS-B, it was calculated that only 15 t ha<sup>-1</sup> of SMS-B need be applied to match the recommended CF application (N: 200 kg ha<sup>-1</sup>, P<sub>2</sub>O<sub>5</sub>:100 kg ha<sup>-1</sup>, K<sub>2</sub>O: 100 kg ha<sup>-1</sup>) in the Kunigami Marge (acidic) soil in Okinawa Prefecture (Department of Agriculture, Forestry and Fisheries in Okinawa Prefecture 2014). However, the available nitrogen content, which is crucial for sugarcane cultivation, may depend on the type of applied organic matter and the use of CFs (Koga et al. 2023, Mochizuki et al. 2023). Therefore, long-term experiments should be conducted to determine whether SMS-B can replace CFs.

Although the carbon concentrations of SMS-B were lower than those of the bagasse and SMS-S, the total soil carbon content in the SMS-B treatment was higher than in both the bagasse and SMS-S treatments (Tables 2, 6). An increase in the soil carbon content after SMS application has been reported in previous studies. For example, Oda et al. (2014) showed a 2.8-fold increase in carbon concentration in surface soils under 4.5 years of SMS application when compared with conventional farming, while Carpio et al. (2023) showed that repeated

application of SMS caused the organic carbon content in surface soil (0 cm-30 cm) to be 2.3 times as much as the initial content in silty loam soil.

The relationship between the amount of applied carbon and the total soil carbon content after the experiment showed that SMS-B retained more carbon than the other treatments (Fig. 2). As noted above, SMS-B had a lower C:N ratio than SMS-S owing to the use of composted bagasse. In general, composting results in a significant decrease in hemicellulose and cellulose and a relative increase in lignin compared with the original crop residue (Kyuma 1997). Bernal et al. (1998) conducted incubation experiments and found that composted organic materials generally minimize carbon mineralization. They also reported that organic materials mixed with more than 50% sweet sorghum bagasse showed the lowest amount of carbon mineralization (38.0% of total carbon). Investigating the effect of composted organic material application on soil organic matter, Rivero et al. (2004) demonstrated in a three-year field experiment that applying composted sewage sludge to acidic soils increased their humic acid content, which is relatively stable against decomposition. Furthermore, previous studies have shown that applying SMS increases the amount of humic acid in the soil (Li et al. 2020). These results indicate that SMS-B may be resistant to degradation in the early stages of sugarcane growth and may also remain stable in the soil over the long term. However, because this pot experiment was conducted over a short period, the carbon sequestration effect of the SMS-B application should be verified through future long-term experiments.

### Conclusions

The application of SMS-B was shown to be effective in increasing the dry matter weight of sugarcane and improving soil chemical properties. This may be due to the higher nitrogen, phosphate, and potassium content and lower C:N ratio than those of bagasse and SMS-S. Furthermore, the application of SMS-B increased the dry matter weight of sugarcane even when chemical fertilizers (CF) were reduced. In addition, SMS-B had a higher soil total carbon content relative to the amount of applied carbon than bagasse and SMS-S. This suggests that applying SMS-B promotes the early growth of sugarcane and effectively reduces its environmental impact.

### Acknowledgements

Nangoku Mushroom Garden Co., Ltd. and Sakura Mushroom Garden Co., Ltd. provided SMS-B and



SMS-S. The bagasse and sugarcane seedlings were provided by Ishigakijima Sugar Co. We thank all those who provided us with materials. The authors thank Yumi Hamada and Wang Jiashuo for their help with soil chemical analysis and data processing. The authors thank the soil analysis team in the Crop, Livestock and Environment Division, JIRCAS, for conducting the soil chemical analyses. We are also grateful to the technical support section of the Tropical Agriculture Research Front, JIRCAS for managing the pot experiments.

## References

- Atallah, E. et al. (2021) Hydrothermal carbonization of spent mushroom compost waste compared against torrefaction and pyrolysis. *Fuel Process. Technol.*, **216**, 106795. <https://doi.org/10.1016/j.fuproc.2021.106795>
- Bijla, S. & Sharma, V. P. (2023) Status of mushroom production: Global and national scenario. *Mushroom Res.*, **32**, 91-98. <https://doi.org/10.36036/MR.32.2.2023.146647>
- Bernal, M. P. et al. (1998) Carbon mineralization from organic wastes at different composting stages during their incubation with soil. *Agric. Ecosyst. Environ.*, **69**, 175-189. [https://doi.org/10.1016/S0167-8809\(98\)00106-6](https://doi.org/10.1016/S0167-8809(98)00106-6)
- Carpio, M. J. et al. (2023) Changes in vineyard soil parameters after repeated application of organic-inorganic amendments based on spent mushroom substrate. *Environ. Res.*, **221**, 115339. <https://doi.org/10.1016/j.envres.2023.115339>
- Chen, L. et al. (2022) Short-term responses of soil nutrients, heavy metals and microbial community to partial substitution of chemical fertilizer with spent mushroom substrates (SMS). *Sci. Total Environ.*, **844**, 157064. <https://doi.org/10.1016/j.scitotenv.2022.157064>
- Committee on Soil Environmental Analysis Methods (1997) Kakyuutai chisso (Available nitrogen). In Committee on Soil Environmental Analysis Methods (ed.), Dojou kankyou bunsekihou (Soil Environmental Analysis Methods). Hakuyusha, Tokyo, Japan, pp. 255-262 [In Japanese].
- Committee on Soil Nutrient Analysis Methods (1970) Bray hou (Bray methods). In Committee on Soil Nutrient Analysis Methods (eds.), Dojou yobun bunsekihou (Soil Nutr. Anal. Methods). Youkendo, Tokyo, Japan, pp. 245-249 [In Japanese].
- Department of Agriculture, Forestry and Fisheries in Okinawa Prefecture (2014) Satoukibi saibai shishin (Sugarcane cultivation guidelines). Department of Agriculture, Forestry and Fisheries in Okinawa Prefecture. [https://www.pref.okinawa.jp/\\_res/projects/default\\_project/\\_page\\_001/010/377/02kanril.pdf](https://www.pref.okinawa.jp/_res/projects/default_project/_page_001/010/377/02kanril.pdf). Accessed on 5 April 2022 [In Japanese].
- Forest Resources Research Center, Department of Agriculture, Forestry and Fisheries, Okinawa Prefecture (2022) Kaitei kinshou shiitake saibai no shishin (Revised guidelines for cultivation of shiitake mushrooms on mushroom beds). Department of Agriculture, Forestry and Fisheries in Okinawa Prefecture. [https://www.pref.okinawa.lg.jp/\\_res/projects/default\\_project/\\_page\\_001/029/487/kaitei\\_shiitake\\_shousai02.pdf](https://www.pref.okinawa.lg.jp/_res/projects/default_project/_page_001/029/487/kaitei_shiitake_shousai02.pdf). Accessed on 10 June 2024 [In Japanese].
- Gobbi, V. et al. (2018) Specific humus systems from mushrooms culture. *App. Soil Ecol.*, **123**, 709-713. <https://doi.org/10.1016/j.apsoil.2017.10.023>
- Hira, M. (2020) Agricultural land in Okinoerabu - Agricultural and rural development for the promotion of island areas: Securing agricultural water and construction of underground dam in Okinoerabu Island, Kagoshima Prefecture. *Suido no Chi (Water Land Environ. Eng.)*, **88**, 595-597 [In Japanese].
- Inoue, K. & Hashiguchi, K. (2011) Effects of bagasse compost applied around planted stalks in a dark-red soil on soil water content and growth of spring-planted sugarcane. *Nihon dojou hiryuu gaku zasshi (Jpn. J. Soil Sci. Plant Nutr.)*, **82**, 398-400 [In Japanese].
- Jayaraman, S. et al. (2024) Mushroom farming: A review focusing on soil health, nutritional security and environmental sustainability. *Farm. Syst.*, **2**, 100098. <https://doi.org/10.1016/j.farsys.2024.100098>
- Jiang, H. et al. (2017) Characteristics of bio-oil produced by the pyrolysis of mixed oil shale semi-coke and spent mushroom substrate. *Fuel*, **200**, 218-224. <https://doi.org/10.1016/j.fuel.2017.03.075>
- Koga, N. et al. (2023) A statistical model with acid detergent-soluble organic nitrogen content of organic amendments predicts nitrogen mineralization of various organic amendments in soil. *Nihon dojou hiryuu gaku zasshi (Jpn. J. Soil Sci. Plant Nutr.)*, **94**, 106-114 [In Japanese with English summary].
- Kuba, M. et al. (1989) Effects of lime and bagasse compost application on soil acidity, exchangeable aluminum, fertility and growth of sorghum in Kunigami-Marge sub soil. *Okinawaken nougyo shikenjou kenkyuu houkoku (Bull. Okinawa Agric. Exp. Stn.)*, **13**, 111-126.
- Kumada, K. (1977) Yuukibutsu no bunkai to fushokuka - fushoku to edaphology (Decomposition and humification of organic matter - Humus and edaphology). In Kumada, K. (ed.), Dojou yuukibutsu no kagaku (Chemistry of Soil Organic Matter). Japan Scientific Societies Press, Tokyo, Japan, pp. 249-276 [In Japanese].
- Kyuma, K. (1997) Dojou yuukibutsu no bunkai to C/N hi (Decomposition of soil organic matter and C:N ratio). In Kyuma, K. (ed.), Saishin Dojou-gaku (Latest Soil Science). Asakura shoten, Tokyo, Japan, pp. 45-46 [In Japanese].
- Leong, Y. K. et al. (2022) Valorization of spent mushroom substrate for low-carbon biofuel production: Recent advances and developments. *Bioresour. Technol.*, **363**, 128012. <https://doi.org/10.1016/j.biortech.2022.128012>
- Li, F. et al. (2020) Spent mushroom substrates affect soil humus composition, microbial biomass and functional diversity in paddy fields. *App. Soil Ecol.*, **149**, 103489. <https://doi.org/10.1016/j.apsoil.2019.103489>
- Miyagi, K. et al. (2002) Characteristics of sugarcane new cultivar Ni15 with early maturity and high quality. *Nihon sakumotsu gakkai kyuushuu shibu kaihou (Rep. Kyushu Br. Crop Soc. Jpn.)*, **68**, 47-49 [In Japanese].
- Miyazato, K. (1986) Bagasu (Bagasse). In Miyazato, K. (ed.), Satoukibi to sono Saibai (Sugarcane and its cultivation). Nihon Bunmitsutou kougyou-kai (Japan Molasses Sugar Industry Association), Naha, Japan, pp. 336-337 [In Japanese].
- Mochizuki, K. et al. (2023) Field validation of a model to

- predict nitrogen mineralization of organic amendments in soil. *Nihon dojou hiryou gaku zasshi (Jpn. J. Soil Sci. Plant Nutr.)*, **94**, 179-186 [In Japanese with English summary].
- Nakanishi, Y. (2017) Effective fertilizer application in sugarcane cultivation to reduce eutrophication and acidification of coral reef seas. *Nihon Sangoshou gakkai-shi (J. Jpn. Coral Reef Soc.)*, **19**, 109-118 [In Japanese with English summary].
- Nakanishi, Y. et al. (2022) Digestibility of fiber components of spent mushroom substrate of *Auricularia nigricans* made from bagasse. *Nihon Danchi Chikusan Gakkai-hou (J. Warm Reg. Soc. Anim. Sci.)*, **65**, 33-36 [In Japanese].
- Nakatsuka, H. et al. (2016) Effects of fresh spent mushroom substrate of *Pleurotus ostreatus* on soil micromorphology in Brazil. *Geoderma*, **269**, 54-60. <https://doi.org/10.1016/j.geoderma.2016.01.023>
- Oda, M. et al. (2014) Application of high carbon:nitrogen material enhanced the formation of the soil A horizon and nitrogen fixation in a tropical agricultural field. *Agric. Sci.*, **5**, 1172-1181. <http://dx.doi.org/10.4236/as.2014.512127>
- Pérez-Chavez, A. M. et al. (2019) Mushroom cultivation and biogas production: A sustainable reuse of organic resources. *Energy Sustain. Dev.*, **50**, 50-60. <https://doi.org/10.1016/j.esd.2019.03.002>
- Rivero, C. et al. (2004) Influence of compost on soil organic matter quality under tropical conditions. *Geoderma*, **123**, 355-361. <https://doi.org/10.1016/j.geoderma.2004.03.002>
- Shinzato, Y. et al. (2013) Effects of drainage improvement for sugarcane fields by subsurface drainage and subsoiling. *Nougyou kikai gakkai-shi (J. Jpn. Soc. Agric. Mach.)*, **75**, 426-433 [In Japanese with English summary].
- Soil Association of Japan (2001) Kakyuutai keisan (Available silicate). In Soil Association of Japan (eds.), *Dojou kinou monitoring chousa no tame no dojou, suishitsu oyobi shokubutsu-tai bunseki-hou* (Soil, water and plant analysis methods for soil monitoring studies). Soil Association of Japan, Tokyo, Japan, pp. 81-86 [In Japanese].
- Sugimoto, A. et al. (2003) Desirable traits of sugarcane variety for improvements of cane yields in lower yielding area in Ryukyu archipelago. *Nihon sakumotsu gakkai kyuushuu shibu kaihou (Rep. Kyushu Br. Crop Soc. Jpn.)*, **69**, 63-66 [In Japanese].
- Tarora, K. et al. (2005) Effects of green manure crops on the growth of next summer-planted sugarcane in early stages. *Nihon sakumotsu gakkai kyuushuu Shibu kaihou (Rep. Kyushu Br. Crop Soc. Jpn.)*, **71**, 75-77 [In Japanese].
- Thomaz, E. L. et al. (2022) Soil erosion on the Brazilian sugarcane cropping system: An overview. *Geogr. Sustain.*, **3**, 129-138. <https://doi.org/10.1016/j.geosus.2022.05.001>
- Umor, N. A. et al. (2021) Zero waste management of spent mushroom compost. *J. Mater. Cycles Waste Manage.*, **23**, 1726-1736. <https://doi.org/10.1007/s10163-021-01250-3>
- Valverde, M. E. et al. (2015) Edible mushrooms: Improving human health and promoting quality life. *Int. J. Microbiol.*, 376387. <https://doi.org/10.1155/2015/376387>
- Vuyyuru, M. et al. (2019) Effects of nitrogen fertilization and seed piece applied fungicides on establishment, tiller dynamics, and sucrose yields in successively planted sugarcane. *Agronomy*, **9**, 387. <https://doi.org/10.3390/agronomy9070387>
- Williams, B. C. et al. (2001) An initial assessment of spent mushroom compost as a potential energy feedstock. *Bioresour. Technol.*, **79**, 227-230. [https://doi.org/10.1016/S0960-8524\(01\)00073-6](https://doi.org/10.1016/S0960-8524(01)00073-6)
- Yamauchi, M. et al. (2015) Development of cultivation technique for cloud ear mushroom (*Auricularia polytricha*) using fermented bagasse and brown sugar shochu lees. *Doboku gakkai ronbunshuu G (Kankyuu) (J. Jpn. Soc. Civ. Engineers, Ser. G (Environ. Res.))*, **71**, 229-237 [In Japanese with English summary].
- Yokoi, D. et al. (2005) Improvement of sugarcane production in volcanic ash soil around Mt. Pinatubo, Pampanga province, Philippines – Evaluation of fertilizer effect –. *Nettai nougyou (Jpn. J. Trop. Agric.)*, **49**, 61-69 [In Japanese with English summary].
- Yoshida, K. et al. (2017) Effect of application of molasses on sugarcane growth, yield, and chemical properties in a red–yellow soil on Kitadaito Island. *Nihon dojou hiryou gaku zasshi (Jpn. J. Soil Sci. Plant Nutr.)*, **88**, 509-518 [In Japanese with English summary].
- Yoshimoto, H. et al. (2016) Effect of *Agaricus* spent compost on the productivity and soil environment for low-input cultivation of forage rice. *Nihon kinoko gakkai-shi (Mushroom Sci. Biotechnol.)*, **23**, 173-178.