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REGULAR ARTICLE

Uptake of carbon and nitrogen derived from carbon-13 and nitrogen-15 dual-labeled maize residue compost applied to radish, komatsuna, and chingensai for three consecutive croppings

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Abstract A pot experiment was conducted to determine the effects of the application of ^{13}C (1.256 atom%) and ^{15}N (1.098 atom%) dual-labeled maize residue compost (MRC) on the nitrogen and carbon uptake by radish, komatsuna, and chingensai as compared with the effect of inorganic fertilizer (IF). The vegetables were grown over three consecutive growing seasons over 4 months; compost was applied at the rate of 24 g kg^{-1} soil. Nonlabeled nitrogen fertilizer was applied to the compost treatments in the second and third crops to compare the effects of blends of compost with N fertilizer to fertilizer alone. The N uptake and yield of vegetables were significantly higher with the recommended inorganic N treatment. The vegetables took up significantly ($P<$ 0.05) lower amounts of N from MRC than from IFs during the three cultivations. The values of the N uptake derived by fertilizer application to the plant exhibited significant differences among different vegetables. Nitrogen recovered by komatsuna and chingensai from MRC was 7.3 (6.6%), 2.7 (1.8%), and 2.3, (1.7%) in the first, second, and third crops, respectively. Radish, komatsuna, and chingensai recovered significant amounts of C from MRC in the first and second crops, with negligible C recovery in the third crop. The initial loss of fertilizer C in soil at the first crop indicates that the microbial decomposition decoupled substantial amounts of $^{13}C/^{15}N$-labeled compounds early in plant development, thus giving the microorganisms a preemptive competitive advantage in the acquisition of easily available $^{13}C/^{15}N$-labeled substrates. It is concluded that a combination of compost and inorganic N did not supply sufficient plant-available N to increase vegetables yields or N uptake over those of fertilizer alone. The data suggested that higher productivity of vegetables might be achieved after the accumulation of a certain amount of residual compost N.

Keywords Maize residue compost · ^{13}C · ^{15}N · Radish · Komatsuna · Chingensai

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Introduction

In field vegetable production, high nutrient demands must be satisfied within relatively short periods by the extensive use of inorganic fertilizers (IF), particularly large amounts of nitrogen (N) fertilizer. However, these systems are often associated with problems such as the accumulation of nutrients in soil, elevated leaching of nitrate (NO_3) from agricultural areas leading to eutrophication of surface waters and contamination of ground water (Ritter 1989), and decreased levels of soil organic matter (Hansen et al. 2001). Therefore, organic inputs, such as plant residues and compost, are recommended to improve the soil's physical structure; the use of organic fertilizers decreases the need for IFs and mitigates environmental degradation (Kirchmann and Thorvaldsson 2000).

Maize (*Zea mays* L.) residue is one of the easily available organic materials that is a high-cellulose feedstock that remains on the land and has an extremely low decomposition rate because of its high C/N ratio. Therefore, composting maize residues may be a useful method of producing a stable product with substantial organic matter that produces humus, which can be used as a source of organic materials and facilitates the slow release of nutrients. The decomposition of maize residues has been studied based on N immobilization (Burgess et al. 2002; Green and Blackmer 1995; Hood et al. 2000; Recous et al. 1995). Generally, the N transformation occurring after compost addition in soil has been studied using the ^{15}N stable isotope technique, and this has been described in various studies (Matsushita et al. 2000; Ueno and Yamamuro 2001; Yamamuro et al. 2002). Moreover, the dynamics and availability of N from compost and IF have been compared (Eghball and Power 1999; Stamatiadis et al. 1999). Recently, there have been a few attempts to more closely integrate the studies of C and N transformation after organic matter addition by using the stable isotope ^{13}C (Ebid et al. 2007; Yamamuro et al. 2002). In this study, we compared the C and N uptake by the following three vegetable species: radish, of which the edible portion is the napiform taproot, although the tops can be used as a leaf vegetable; chingensai, a kind of Chinese cabbage which responds better to organic N (Matsumoto et al. 2000); and komatsuna, a fast-growing Japanese leafy vegetable.

In a previous study (Ebid et al. 2007), we assessed the application effect of maize residue compost (MRC) on the fate of C and N in paddy soils used for rice cultivation and reported that rice took up N at the rate of 36.8 mg N pot^{-1}, 6.4% from the MRC. Moreover, rice recovered a considerable amount of carbon (3.70%) from applied MRC with the ^{13}C concentration being highest in straw (2.44% of applied C) as compared with grain (0.33%) and root (0.97%).

The objectives of this study were: (1) to assess the C and N uptake between radish, komatsuna, and chingensai treated with MRC and IF, (2) to quantify the available C and N from MRC to radish, komatsuna, and chingensai plants, and (3) to elucidate the distribution of C and N between soil and plant with time.

Materials and methods

Site and soil characteristics

In 2005, three successive pot experiments were carried out at the university farm, Ehime University, Matsuyama, Ehime, Southwest Japan (lat. 33°57′ N and long. 132°47′ E). Low fertility upland, brown forest soils, classified as a Dystric Regosol (FAO/UNESCO 1987) was used collected from the top 0-20 cm, sieved, and a <2-mm fraction was obtained. The soil properties were as follows: pH (H_2O) 6.60, electrical conductivity (EC) 0.311 dS m^{-1}, total C 1.12%, total N 0.133%, cation exchange capacity (CEC) 16.5 cmol(+) kg^{-1}, C/N 8.4, available phosphorus (P) 1,897 mg kg^{-1}, exchangeable potassium (K) 623 mg kg^{-1}, Ca 1,349 mg kg^{-1}, and Mg 325 mg kg^{-1}. The mean diurnal soil temperature of the pots during the cultivation period was 35/12.7°C (maximum/minimum), and the mean ambient temperature was 22.7°C.

Plant establishment and experimental design

The trial was carried out during the vegetable cropping season from May to September 2005 under greenhouse conditions. Three vegetable species, komatsuna (*Brassica rapa* L. cv. "Rakuten"), radish (*Raphanus sativus* L. cv. "Radicula Pers"), and chingensai (*Brassica campestris* L. cv. "Choyo No. 2"), were raised in plastic pots with dimensions of 83 cm (L) × 27 cm (W) × 20 cm (H) filled with 40 kg of soil (dry weight). The three fertility treatments in five replicates were as follows: (1) control treatment to which additional N was not applied. (2) IF pots to which N fertilizer labeled with 10.5 atom% was applied as ammonium chloride at 3.36 g pot^{-1} (84.4 mg N kg^{-1} dry soil) at only the first cropping. Further, P and K were supplied at a rate of 120 kg ha^{-1} as phosphorus pentoxide (P_2O_5, 13.4 g pot^{-1}) and potassium chloride (KCl, 2.68 g pot^{-1}), respectively. (3) Organic fertilizer pots supplied with ^{13}C (1.256 atom%) and ^{15}N (1.098 atom%) dual-labeled (Yamamuro et al. 2002) MRC [73.0% moisture content, 35.2% T-C and 2.9% T-N (dry matter), and 12.1 C/N]. The compost was applied at a rate of 672.3 g (123.7 g dry matter) pot^{-1}, which corresponded to 3.66 g N pot^{-1} (91.5 mg N kg^{-1} dry soil). The normal organic fertilizer application rate ranges from 300 to 350 kg N ha^{-1} (Kramer et al. 2002; Burger and Jackson 2003) depending on the N requirement of the crop and soil N availability. Three consecutive crops of the three species were cultivated over 4 months from 2 May to 1 September (2005) in their respective pots, and every crop was 40 days old. Nonlabeled N fertilizer was applied to the compost treatments in the second and third crops at half of the quantity applied to IF in each crop for improvement of the short-term effectiveness of MRC. The water content in the pots was maintained between 40 and 80% water-holding capacity (WHC).

Sampling and isotope analysis

The vegetables were harvested at commercial maturity and only the edible portions were used for analysis. Radish plants were separated into leaf and root (succlulent root), while only leaves were sampled for chingensai and komatsuna, then oven dried at 75°C for 42 h. The dried samples were weighed and ground into a fine powder with an electric mill. After harvesting, the soil in the pots was homogenized before sampling and immediately oven dried at 75°C for 4 days. The dried soil subsamples were finely pulverized and then ground. The plant and soil samples were subjected to automatic combustion in tin (Sn) capsules and analyzed by mass spectrometry using a stable isotope mass spectrometer (ANCA-SL, Europa Scientific Co. Ltd., Crewe, UK). The ratios of ^{13}C and ^{15}N to the total C and N content in the samples were determined; all the treatments were carried out in five replicates.

Calculation of C and N dynamics

1. The atom% of ^{13}C and ^{15}N excess was calculated based on the difference between the ^{13}C and ^{15}N atom% in the plants and natural abundance according to the PeeDee belemnite ODB standard (1.11%) and in the atmosphere (0.3663%).
2. The atom% of ^{13}C and ^{15}N was recalculated by subtraction from that of the labeled corn residue compost sample to which nonlabeled corn compost was applied.
3. Derived N (%) = (^{15}N atom% excess of N)/(^{15}N atom% excess of applied N) × 100.
4. Derived N (mg) = (^{15}N atom% excess of N)/(^{15}N atom% excess of applied N) × total N.
5. The recovery of ^{13}C and ^{15}N from soil and plant pools analyzed by mass spectrometry was calculated using the following equation
   Recovery percentage of N and C = ^{15}N or ^{13}C plant (kg ha^{-1} of N or C)/N or C rate (kg ha^{-1} of N or C) × 100.
6. The emission rates of C (or N) from the MRC applied were calculated using the following equation

$$E = 1 - (P + 1)$$

where E denotes emission, P denotes plant uptake, and I is the amount of N remaining in soil (= residual N, immobilization, and assimilation).

Statistical analysis

Statistical analyses were conducted by analysis of variance and Fisher's protected significant difference $P<0.05$ and the differences among the means were analyzed by the Tukey-Kramer test using the software KyPlot (KyensLab Inc., Tokyo, Japan).

Results and discussion

Dry matter production and N uptake

Table 1 shows the dry matter production and total N uptake by the vegetables. In all of the trials, the effect of IF on vegetable dry matter was significantly higher than that of control and MRC treatment. However, no significant difference was observed between all three treatments, control, IF, and MRC in radish leaf dry matter in the first and third crops. The vegetable dry matter in the third crop was lower than in the first and second crops. Total N uptake in the vegetables had a

Table 1 Dry matter yield and total nitrogen (N) uptake by vegetables grown in soil amended with either inorganic fertilizer (IF) or maize residue compost (MRC)

Plant part	Treatment	Dry matter (g pot^{-1})			Total N (mg pot^{-1})		
		1st crop	2nd crop	3rd crop	1st crop	2nd crop	3rd crop
Radish root	Control	3.52a	2.72a	2.37a	90.4a	69.5a	54.9a
	IF	5.75b	4.60b	4.07b	208.6b	187.3b	132.0b
	MRC	5.21c	3.75c	3.03c	173.1c	100.4c	86.9c
	F value	**	**	*	***	**	***
Radish leaf	Control	3.34a	2.16a	2.05a	168.3a	91.9a	68.2a
	IF	3.98a	3.49b	2.76a	208.2b	193.9b	137.1b
	MRC	3.81a	2.92c	2.37a	197.3c	127.6c	112.1c
	F value	NS	**	NS[d]	*	**	***
Komatsuna leaf	Control	12.72a	10.46a	7.38a	549.1a	374.2a	188.8a
	IF	15.74b	14.88b	11.26b	908.2b	856.8b	622.3b
	MRC	14.02c	13.03c	8.24c	636.9c	423.5c	219.9c
	F value	**	**	**	**	***	***
Chingensai leaf	Control	10.07a	5.28a	4.17a	433.4a	122.8a1	19.9a
	IF	13.05b	11.82b	10.92b	723.8b	653.7b	520.8b
	MRC	11.62c	8.15c	6.47c	493.0c	246.8c	181.0c
	F value	*	***	***	**	***	***
	F value[e]						
	Control	***	***	***	***	*	**
	IF	***	***	***	***	***	***
	MRC	***	***	***	***	***	***

Different letters in each column denote significant differences within treatments (Tukey-Kramer test, $P<0.05$, $n=5$)

[d] NS, not significant at $P>0.05$

[e] For comparison between vegetables within the same treatment

similar pattern to dry matter production (Table 1). Total N was significantly higher in the first crop and decreased in the second and third crops.

Different N uptake responses were observed in the vegetable species, with komatsuna acquiring the greatest amount of N followed by chingensai and radish (Table 1). The vegetables responded differently to MRC and IF treatments; N uptake by the vegetables was highest with IF treatment. In this study, we used the harvestable portion of vegetables to provide the evidence for whether compost-fertilizer blends are more efficient in providing N than fertilizer alone. Data indicated that neither yield nor N uptake increased when MRC was blended with N fertilizer. Vegetable yield and the total N uptake did not increase with MRC treatment in agreement with Sikora and Enkiri (2001), who reported that compost blended with N fertilizer did not supply sufficient additional available N to increase yields.

Moreover, Azam et al. (1985) reported that the fertilizer N use efficiency (NUE) decreased when organic and inorganic sources of N were combined. However, non-isotope studies have indicated that some blends of compost and fertilizer enable yields and N uptake greater than those obtained by fertilizer treatments alone (Sikora and Enkiri 1999).

^{15}N recovery and N derived from fertilizers (Ndff) and soil (Ndfs)

N uptake derived from MRC was detected in all of the vegetable species in the first, second, and third crops (Table 2). N uptake preliminarily derived from IF treatment was significantly higher than that with MRC treatment. N derived from the IF applied at the first cropping decreased gradually in the following order: first crop > second crop > third crop. However, with radish leaf and root, N derived from MRC followed the trend first crop > third crop > second crop. N uptake with IF or MRC treatment in radish leaf was higher than in radish root. However, with chingensai leaf, no significant difference was ob-

Table 2 Uptake of N derived from inorganic fertilizer (IF) or maize residue compost (MRC), N derived from soil, and percentage of ^{15}N recovered in vegetables; calculations performed using the direct method. *Ndff* N derived from fertilizer (IF and MRC), *Ndfs* N derived from soil calculated by total N uptake by plants − N derived from IF or MRC, *NRc* percentage of ^{15}N recovery in vegetables

Plant part	Treatment	1st crop			2nd crop			3rd crop		
		Ndff (mg pot^{-1})	Ndfs	NRc %	Ndff (mg pot^{-1})	Ndfs	NRc %	Ndff (mg pot^{-1})	Ndfs	NRc %
Radish root	Control	–	90.4a	–	–	69.5a	–	–	54.9a	–
	IF	109.0a	99.6a	9.8a	37.9a	149.4b	3.4a	24.5a	107.5b	2.2a
	MRC	22.3b	150.8b	1.8b	8.4b	92.0c	0.7b	12.6b	74.3c	1.1b
	F value	***	**	**	**	***	**	**	***	*
Radish leaf	Control	–	168.3a	–	–	91.9a	–	–	68.2a	–
	IF	111.0a	97.2b	9.9a	40.9a	153.0b	3.7a	21.1a	116.0b	1.9a
	MRC	28.0b	169.3a	2.3b	12.7b	114.9c	1.1b	14.4b	97.7c	1.2a
	F value	***	***	*	**	***	*	**	**	NS
Komatsuna leaf	Control	–	549.1a	–	–	374.2a	–	–	188.8a	–
	IF	485.6a	422.6b	43.3a	160.7a	696.1b	14.4a	106.9a	515.4b	9.5a
	MRC	89.4b	547.5a	7.3b	33.1b	390.4a	2.7b	27.3b	192.6a	2.3b
	F value	***	*	*	**	**	**	*	*	**
Chingensai leaf	Control	–	433.4a	–	–	122.8a	–	–	119.9a	–
	IF	344.7a	379.1a	30.8a	139.3a	514.4b	12.4a	82.8a	438.0b	7.4a
	MRC	80.0b	413.0a	6.6b	21.6b	225.2c	1.8b	20.5b	160.5c	1.7b
	F value	***	NS	**	***	*	**	***	**	**
	F value									
	Control	–	***	–	–	***	–	–	***	–
	IF	***	***	***	***	***	***	***	***	***
	MRC	***	***	***	***	***	***	***	***	***

Different letters in each column denote significant differences within treatments (Tukey-Kramer test, $P<0.05$, $n=5$)

served between N uptakes derived from MRC treatment in the second and third crops.

In this study, under upland conditions, the amounts of inorganic N derived from the soil at the first, second, and third croppings was significantly different between IF and MRC treatments (Table 2). For all crops, the amount of N derived from soil treated with MRC was lower than that derived from soils treated with IF in the second and third crops. However, for the first crop the amount of N derived from the soil with MRC treatment was significantly higher than that derived by IF soil treatment.

In this study, a significant amount of N uptake by vegetables was observed, although the rates varied with the plants; especially komatsuna and chingensai took up a higher amount of N from MRC than radish.

The amount of inorganic N in the added compost N increased at the first cropping but decreased at the second cropping because the uptake of inorganic N by plants was faster than the mineralization rate of compost after the fraction that easily mineralized was done. Generally, it is considered that the initial mineralization of organic N from MRC in the first cropping is probably due to microbial decomposition of the easily decomposable portion of the MRC, which is generally heterogeneous and consists of more than two components that decompose at different rates (Hadas et al. 1996). Also, as suggested by Kuzyakov et al. (2000), the release of inorganic N without a lag phase is common after the addition of a readily available organic substance because the microbial community does not have to adapt to the added substance.

Based on the low relative efficiency of MRC, it is observed that N from compost alone would not provide sufficiently high yields with short-term applications. Benitez et al. (1998) and Hadas et al. (1996) reported that the short-term availability of N from organic amendments was much lower than that from inorganic inputs due to the low mineralization potential of organic fertilizers. However, higher productivity might be achieved after the accumulation of a certain amount of residual MRC N as reported by Sikora and Scott (1996), indicating that the influence of organic matter addition to soils is evident only after several years of application. Tester (1990) reported that when compost application rates were greater than 50 mg ha^{-1}, the organic matter content of soils was significantly increased and the soil's physical properties were affected.

In this study, komatsuna and chingensai recovered a much larger amount of N from MRC in the first crop (7.3, 6.6%) than in the second (2.7, 1.8%) and third crops (2.3, 1.7%). Yamamuro et al. (2002) reported identical levels of N recovery by rice and corn plants from cattle manure compost (17.2 and 10% N, respectively). However, in our previous study (Ebid et al. 2007), we reported that rice plants recovered N from MRC at a rate of 26.8% during 5 months of cultivation.

Carbon uptake by vegetables

Table 3 shows the total carbon uptake by vegetables from IF and MRC. There was no significant difference between total C content in the first crop between IF and MRC; however, in the second and third crops the percentage of total carbon uptake from MRC was significantly higher than the uptake from IF. Total C content in the radish leaf and root increased gradually with cultivation time. However, in komatsuna and chingensai, it increased at the second cropping but decreased at the third cropping (Table 3). The total C content was higher in radish root than other vegetables. At the second cropping, komatsuna had a higher total C uptake followed by radish root, and at the third cropping, radish leaf had a higher total C uptake followed by radish root. The final recovery rate of ^{13}C in the first cropping was significantly higher in chingensai and radish leaf; however, in the second cropping, it was significantly higher in chingensai and komatsuna, and in the third cropping there was no significant difference between the vegetables. The results demonstrate that the recovery rates of C from MRC varied with the plant species. In particular, chingensai took up a much larger amount of C from MRC than radish and komatsuna (Fig. 1).

Carbon uptake derived from MRC sharply decreased in the second crop of the radish leaf, but the total C content tended to be higher in the second crops than in the first crops of chingensai, komatsuna, and radish root. However, derived C was generally low in the third crop (Fig. 1). Yamamuro et al. (2002) also have suggested that rice and corn plants absorb C from cattle manure compost at a rate of 2.16 and 13%, respectively. Moreover, we reported that rice took up

Table 3 Total C and ^{13}C recovered in vegetable tissue expressed as a % of initial organic carbon content of inorganic fertilizer (IF) and labeled maize residue compost (MRC)

Vegetable species	Treatment	1st crop		2nd crop		3rd crop	
		T-C (%)	^{13}C recovery (%)	T-C (%)	^{13}C recovery (%)	T-C (%)	^{13}C recovery (%)
Radish root	IF	34.6	–	35.2	–	34.7	–
	MRC	35.4	0.102	36.2	0.133	36.9	0.068
	F value	NS[a]	–	*	–	**	–
Radish leaf	IF	31.8	–	30.5	–	32.9	–
	MRC	33.1	0.239	34.9	0.066	37.3	0.050
	F value	NS	–	**	–	**	–
Komatsuna leaf	IF	31.5	–	33.8	–	31.7	–
	MRC	33.6	0.175	38.1	0.203	34.6	0.019
	F value	NS	–	*	–	*	–
Chingensai leaf	IF	28.9	–	32.4	–	30.2	–
	MRC	33.5	0.275	34.9	0.352	32.7	0.056
	F value	NS	–	**	–	**	–
F[b] value	IF	*	–	***	–	***	–
	MRC	NS	*	***	NS	NS	NS

[a] NS, not signif8icant at $P>0.05$

[b] For comparison between vegetables

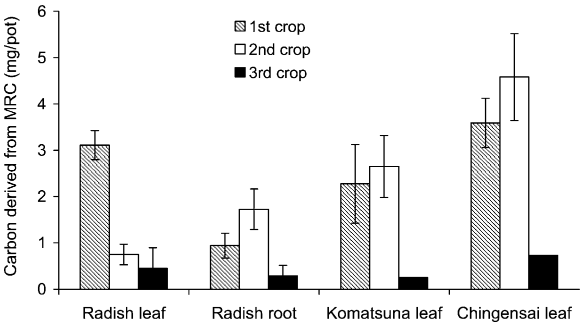

Fig. 1 Carbon uptake by radish, komatsuna, and chingensai derived from maize residue compost (MRC) in the 1st, 2nd, and 3rd crops. *Vertical bars* represent ± standard error (SE, n=5)

C from MRC at a rate of 3.7% (Ebid et al. 2007). Further, Chidthaison and Watanabe (1997) reported that rice plants recovered 1.0% of the C from ^{13}C-labeled rice straw.

^{15}N and ^{13}C distribution from MRC

Table 4 shows the distribution of ^{15}N and ^{13}C from MRC. Nitrogen emission from MRC after the first crop was very low (4.0%) and then gradually increased after the second (9.7%) and third crops (11.7%). On the other hand, carbon emission from MRC sharply increased after the first crop (48.1%) and then gradually decreased after the second (17.3%) and third crops (12.0%). The percentage of remaining N from MRC was 78% after the first crop and then decreased gradually and reached values of 60.9 and 42.9% at the second and third crops, respectively. The N losses were low because the water content in the pots was maintained between 40 and 80% WHC. NH_3 losses may be less than 1–4% of the total plant N as observed by Schjoerring and Mattsson (2001). Also, Rroco and Mengel (2000) reported that the ^{15}N losses were between 3.6 and 4.7% in a pot experiment with wheat performed using the ^{15}N prelabeling method.

A considerable difference was observed in the patterns of C and N mineralizations in agreement with Nelson and Oades (1996) and Yamamuro et al. (2002). Trinsoutrot et al. (2000) demonstrated that the rapid increase in the C release was caused by the exponential growth of soil aerobic microbes that utilized easily decomposable and metabolizable C compounds such as soluble organic matter. Several studies (Gilmour et al. 1985; Hadas and Portnoy 1994; Molina et al. 1983) found a significant correlation between C evolution from CO_2 and inorganic N release from composts.

Conclusions

The results of the present study revealed little positive short-term effect of MRC application on vegetable N uptake and N mineralization under upland conditions due to the low contribution of MRC-derived N to vegetables and soil inorganic N. However, the effect varied with vegetable species, suggesting long-term incorporation of MRC may contribute to improve soil fertility, growth, and N uptake of vegetables. Further studies are necessary to clarify the plant nutrient uptake and nutrient dynamics in soils to evaluate the effect of organic matter application in soil and developing “best management practices” for compost as a partial alternative to IF for commercial vegetable production.

Table 4 Comparison of ^{15}N and ^{13}C distribution from MRC at vegetable harvest

	Distribution (%)		
	Plant uptake	Remains in soil	Emission
Nitrogen			
First crop	18.0a	78.0a	4.0a
Second crop	7.4b	60.9b	9.7b
Third crop	6.3b	42.9c	11.7c
Carbon			
First crop	0.791a	51.2a	48.1a
Second crop	0.754a	33.1b	17.3b
Third crop	0.193b	20.0b	12.0b

Values with the same letter in each column indicate no significant differences (Tukey-Kramer test, $P<0.05$, n=5)

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