Soil carbon and nitrogen and tomato yield response to cover crop management

Rafael A. Muchanga\textsuperscript{1,4} | Toshiyuki Hirata\textsuperscript{2} | Yoshitaka Uchida\textsuperscript{3} | Ryusuke Hatano\textsuperscript{3} | Hajime Araki\textsuperscript{2}

\textsuperscript{1}Graduate School of Environmental Science, Hokkaido University, Sapporo, 060–0810, Japan
\textsuperscript{2}Field Science Center for Northern Biosphere, Hokkaido University, Sapporo, 060–0811, Japan
\textsuperscript{3}Research Faculty of Agriculture, Hokkaido University, Sapporo, 060–8589, Japan
\textsuperscript{4}Current address: Faculty of Agronomic Engineering and Forestry, Zambeze University, Mocuba, 2403, Mozambique

Correspondence
Hajime Araki, Field Science Center for Northern Biosphere, Hokkaido University, Sapporo 060–0811, Japan
Email: araki@fsc.hokudai.ac.jp

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Abstract
Depending on management, cover crops may improve soil and environmental quality and tomato (\textit{Solanum lycopersicum} L.) yield. We evaluated the effects of hairy vetch (HV; \textit{Vicia villosa} R.) residue management and the biculture of HV and rye (\textit{Secale cereale} L.) on soil organic carbon (SOC) and total nitrogen (STN), microbial biomass nitrogen (MBN), soil inorganic nitrogen, and tomato yield for 2 yr in a plastic high tunnel. The SOC in the surface 10-cm depth was 2.87–17.5\% significantly greater in HV incorporation (HVI), HV mulch (HVM), and the biculture of HV and rye treatments (HV\textsuperscript{+}RYE), than in a no cover crop treatment (bare fallow). However, cover crop management effects on STN varied with soil depths (0- to 10- and 10- to 30-cm depths) and years, and HVI tended to be more effective than other treatments in increasing STN. Residual soil nitrate\textsuperscript{−}N was increased by cover crops, more so by HV monoculture than HV\textsuperscript{+}RYE. The MBN and inorganic N (NO\textsubscript{3}\textsuperscript{−}N + NH\textsubscript{4}\textsuperscript{+}N) were greater in HVI than either HVM or HV\textsuperscript{+}RYE. Tomato total yield was 11.1–43.8\% significantly greater in HVI and HVM than in bare fallow. However, the effects of HV\textsuperscript{+}RYE on MBN, inorganic N, and tomato yield varied with C/N ratio of residues and best results were obtained with a C/N ratio of 17.6 than with 23.7. Therefore, if an adequate seeding HV\textsuperscript{+}rye ratio (2:1) is used, HV\textsuperscript{+}RYE is a better management practice to increase SOC and STN at topsoil and tomato yield with least residual N.

1 | INTRODUCTION

The use of winter cover crops in crop production has emerged as a promising practice to help improve soil quality and crop productivity (Kuo, Huang, & Bembenek, 2001). Winter cover crops use residual soil nitrate–N that may leach into groundwater after crop harvest, thereby reducing the fertilizer inputs (Hargrove, 1986; Kuo, Sainju, & Jellum, 1997a, 1997b; Meisinger, Hargrove, Mikkelsen Jr, Williams, & Benson, 1991; Sainju, Singh, & Whitehead, 2000). The quantity, quality, and management of plant residues are key factors influencing soil carbon (C) and nitrogen (N) pools and crop yields. The quality of residues (N content and C/N ratio) influences N uptake and crop productivity through controls of N mineralization and immobilization (Paustian, Parton, & Persson, 1992). Hairy vetch (HV; \textit{Vicia villosa} R.), with a low C/N ratio, showed greater soil N availability to corn (\textit{Zea mays} L.) compared to other cover crops (Hoskins, 1999). The factors affecting C and N pools in the soil include the amount of organic C in the plant residues, the C/N ratio of residues, and decomposition rates (Paustian, Parton, & Persson, 1992).
mohua (Secale cereale L.) followed by the bicultures of HV and rye (Secale cereale L.) or ryegrass (Lolium multiflorum Lam.). However, rye or ryegrass had either no effect or adverse effect on soil N availability (Kuo et al., 2001). Thus, nonlegumes, such as rye, do not add N to cropping systems but help conserve N through crop uptake and immobilization (Meisinger et al., 1991; Shiplely, Meisinger, & Decker, 1992). Although both legume and nonlegume cover crops can add organic matter to soils, nonlegumes may be more effective than legumes in increasing soil C and N because of greater biomass production (Kuo et al., 1997a, 1997b; Sainju, Whitehead, & Singh, 2003).

Biculture of HV and rye can be an effective management practice to lower C/N ratio of the mixture (Sainju, Whitehead, & Singh, 2005), thereby reducing its potential for N immobilization. Also, the biculture may add more C to the soil than HV (Clark, Decker, & Meisinger, 1994; Sainju et al., 2005). Moreover, the placement of cover crop residues in the soil (incorporation or mulch) may also affect soil C and N pools differently and enhance tomato (Solanum lycopersicum L.) productivity. Generally, decomposition rates of plant residues that are incorporated in the soil are faster than when residues are placed on the soil surface (Chen, Liu, Tian, Yan, & Zhang, 2014; Coppens, Garnier, Findeling, Merckx, & Recous, 2007). Because of slow decomposition of surface-placed residues, the net release of N is delayed (Bradford & Peterson, 2000). However, research on cover crop use in vegetable production in northern Japan, which is characterized by short summer and long winter with heavy snowfall, is still limited. Therefore, the effects of HV residue management (incorporation or mulch) and biculture of HV and rye on soil quality and fresh-market tomato yield are not well-known. Hairy vetch and rye cover crops were used in this study because of their ability to overwinter in northern Japan.

The soil inorganic N (NO$_3^-$–N + NH$_4^+$–N) after crop harvest in fall may represent 50–80% of the total N applied by synthetic fertilizer (Di & Cameron, 2002). The potential for N loss through nitrate leaching may be high in vegetable production systems in northern Japan, especially after winter when snowmelt begins. Therefore, assessment of residual N following cover crop application is important for establishing sustainable tomato production systems. The objective of this study was to evaluate the effects of HV residue management and biculture of HV and rye on fresh-market tomato yield, soil organic C (SOC) and soil total N (STN), microbial biomass N (MBN), and soil inorganic N. Our research hypotheses were: (a) that because of increased biomass production, the biculture of HV and rye may be more effective than HV monoculture, regardless of residue management, in increasing SOC and STN; and (b) because of faster decomposition, HV incorporation would increase tomato yield and residual N more than the biculture and HV mulch.

### Core Ideas
- Cover crop residue management influenced differently soil carbon and nitrogen pool and tomato yield.
- Regardless of management, cover crop treatments increased slightly soil organic carbon at surface 10 cm depth compared with bare fallow.
- Incorporation of hairy vetch residues was the most effective management in increasing soil organic carbon, soil nitrogen availability, and microbial biomass nitrogen.
- Residual nitrate-nitrogen increased with hairy vetch and rye monocultures more than with the biculture of hairy vetch and rye.
- The effects of the biculture on tomato yield, microbial biomass nitrogen, and soil nitrogen availability varied with carbon/nitrogen of residues and the best results were obtained with a carbon/nitrogen of 17.6 than with 23.7.

### 2 MATERIALS AND METHODS

#### 2.1 Site, experimental design, and treatments

Two experiments were conducted in different plastic high tunnels in 2017 and 2018 at the Experimental Farm of the Field Science Center for Northern Biosphere (43°4’N 141°20’E), Hokkaido University, Sapporo, Japan, on a Gleysol with a clay loam texture. Sapporo has a Köppen climate regime of Dfa (humid continental climate), with an average air temperature (30-yr average) ranging from ~3.6°C in January to 22.3°C in August, and annual precipitation of 1,106 mm. The plastic high tunnels (Site 1 [2017], 14.5 by 6.5 by 3.5 m; Site 2 [2018], 20 by 6.5 by 3.5 m) were about 300 m apart, and the chemical properties of the soil at the two sites are presented in Table 1. To determine the initial soil chemical composition, samples were collected from the 0- to 10-cm soil depth at each site, air-dried, sieved with 2-mm mesh, and then analyzed for P$_2$O$_5$, CaO, K$_2$O, MgO, NH$_4^+$–N, and NO$_3^-$–N calorimetrically with a soil analyzer (ZA-II; Fujihiro Industry, Tokyo, Japan). Electrical conductivity (EC) and pH were analyzed from 1:5 (w/v) and 1:2.5 (w/v) extracts, respectively. The soil and air temperatures measured during the period of tomato cultivation are presented in Figure 1. The temperature within the plastic high tunnel was measured in 2017 only with a data logger (Logger GL820; Graphitec, Yokohama, Japan), while the temperature outside the plastic high tunnel was measured in both years by a weather station located at Experimental Farm of Field Science Center for Northern Biosphere, Hokkaido University, Sapporo. Soil and
FIGURE 1 Mean soil and air temperature measured hourly during the period of tomato cultivation (a) inside and outside the plastic high tunnel in 2017 and (b) outside the plastic high tunnel in 2018. Soil and air temperatures (inside and outside the high tunnel) were measured at a depth of 10 cm and a height of 1.5 m.

TABLE 1 Soil chemical properties measured at 0 to 10 cm depth before the initiation of the experiments in 2017 (Site 1) and 2018 (Site 2)

| Property             | Site 1 | Site 2 |
|----------------------|--------|--------|
| pH                   | 5.85   | 5.80   |
| EC, mS m⁻¹           | 31.5   | 42.1   |
| Total C, g kg⁻¹      | 31.3   | 53.1   |
| Total N, g kg⁻¹      | 2.18   | 3.42   |
| C/N                  | 14.2   | 15.5   |
| \( \text{NO}_3^- \) - N, mg kg⁻¹ | 66.3  | 24.2   |
| \( \text{NH}_4^+ \) - N, mg kg⁻¹ | 27.5  | 48.0   |
| P, mg kg⁻¹           | 170    | 60.0   |
| K, mg kg⁻¹           | 190    | 360    |
| Ca, mg kg⁻¹          | 1,710  | 1,460  |
| Mg, mg kg⁻¹          | 400    | 370    |

In 2017 (Site 1), the treatments consisted of bare plots (BARE−F, no cover crop with N fertilization), hairy vetch incorporation (HVI), hairy vetch mulch (HVM), and the biculture of hairy vetch and rye (HV+RYE; incorporation). All plots received a supplemental N fertilizer applied at a rate of 150 kg N ha⁻¹ as LPS100 (41% N; JCAM AGRI, Tokyo, Japan; 30-d-delay in N release in the soil at 25°C, and then 70-d release of 80% of its total N following a sigmoidal pattern). In 2018 (Site 2), the treatments were bare plots without N fertilization (BARE−NF) and BARE-F (N fertilizer applied as LPS100), HV incorporation (HVI), HV mulch (HVM), biculture of HV and rye (HV+RYE), and rye monoculture (RYE). As in 2017, all cover crop plots, received a supplemental N fertilizer applied at a rate of 150 kg N ha⁻¹ as LPS100. This fertilizer amount is equivalent to about 50% of the recommended N fertilizer amount for fresh-market tomato production across different regions in Japan. The treatments were arranged in a randomized complete block design with three replicates. The plots were 2.5-m long and
1-m wide in both years. Each plot contained 10 plants in double lines spaced 0.5 m between plants and 0.5 m between lines, only six plants in the core plot were tested. The distance from the center to center between beds was 1.5 m in both years, resulting in a planting density of 26,667 plants ha\(^{-1}\).

### 2.2 Field preparation and cover crops cultivation

At both sites, the soil was tilled by a rotary cultivator in fall and beds 15-cm high were raised. At Site 1, HV ‘Kantaro’ (overwintering cultivar; Snow Brand Seed, Hokkaido, Japan) was sown at a seeding rate of 20 kg ha\(^{-1}\) (47 seeds m\(^{-2}\)) in the monoculture and biculture plots on 6 October 2016 by hand, and rye ‘Fuyumidori’ (HOKUREN Federation of Agricultural Cooperatives, Hokkaido, Japan) was sown at a seeding rate of 10 kg ha\(^{-1}\) (36 seeds m\(^{-2}\)). At Site 2, HV ‘Kantaro’ was planted at a seeding rate of 50 kg ha\(^{-1}\) in both monoculture and biculture plots on 21 September 2017. Rye was planted at seeding rates of 100 kg ha\(^{-1}\) and 50 kg ha\(^{-1}\) in monoculture and biculture plots, respectively. The seeding rate of HV used at Site 1 (first growing season) was based on the results from Muchanga, Hirata, & Araki (2017), and the high tunnel remained covered with a plastic during the winter because its structure was robust to withstand to heavy snowfall. However, at Site 2, the seeding rates were increased because the planting density of cover crops was expected to be reduced by snow that usually accumulates during the winter, and plastic cover of the tunnel was removed during the winter. The seeding HV/rye ratio (in kg ha\(^{-1}\)) was 2:1 in the first season and 1:1 in the second season.

However, the HV ‘Kantaro’ planted at Site 2 (second season) did not overwinter because of a long snow cover period and high snowpack. Snowfall started from mid-November 2017 to late March 2018. Because of delayed snow melting, HV ‘Mamesuke’ (nonwinter-hardy; Snow Brand Seed, Hokkaido, Japan) was sown in rice trays (60 by 30 cm) using a seeding density equivalent to 50 kg ha\(^{-1}\) on 9 April 2018, and the HV seedlings were transplanted in the plots on 25 April 2018. In the biculture plots, HV was transplanted within rye rows. No fertilizer or agrochemicals were applied to cover crops to supply additional nutrients or to control pests. Water was applied by an irrigation tube as needed after snow cover at Site 1, and from HV transplanting until termination at Site 2. Cover crops were terminated at the flowering or heading stage by cutting the plants by hand 2–3 cm above the ground on 22 May 2017 at Site 1 and on 17 June 2018 at Site 2. Cover crop residues for incorporation were cut into pieces of about 10-cm. Noncover-crop plots were left bare throughout the growing period of cover crops in both years.

### 2.3 Tomato seedling production, transplanting, and management

Tomato rootstock cultivar ‘Friendship’ (Sakata Seed, Tokyo, Japan) was sown in 128-cell trays (cell size 3 by 3 by 4 cm) on 3 March 2017, while the scion cultivar ‘Reika’ (Sakata Seed, Tokyo, Japan) was sown in plastic boxes on 9 March 2017. Tomatoes were grafted on 13 April 2017 using a tube grafting method, and 3 wk later the grafted seedlings were transferred into plastic pots of 12-cm diameter, where they grew until the emergence of the first flower cluster. On 26 May 2017 at Site 1, 2.5-m-old tomato seedlings were transplanted in the plots by hand.

In 2018, nongrafted tomato seedlings were used because of unsuccessful grafting. Fresh-market tomato ‘Reika’ cultivar was sown in 128-cell trays on 3 April, and then seedlings were transferred into 12-cm diameter pots 3 wk after emergence. Tomato seedlings were transplanted in the plots on 20 June. Fertilizers and HV residues were incorporated by hand 2–3 d before transplanting. The soil in the mulch plots was not tilled after HV termination, so fertilizers were spread on the soil surface and then covered by mulch. Phosphorus and potassium fertilizers were applied as fused calcium magnesium phosphate (29% Ca, 7% Mg, 7% P), and as potassium sulfate (42% K, 17% S) at a rate of 200 kg ha\(^{-1}\) each, respectively. After transplanting, irrigation tubes were installed to water tomatoes as needed until the last harvest in each year. Two tension meters (DIK-8330; Daiki Rika Kyogo, Saitama, Japan) were set in each replicate or block, and pF values were monitored daily. Irrigation was performed when pF >2.3. For trellising tomato plants, a hanging-string system was established just after transplanting. Lateral shoots were removed as they appeared for allowing only the main shoot to grow, and pollination occurred naturally. Fruits were collected from the first to eighth cluster, and only four fruits were allowed to set in each cluster. Pesticides were applied three times to control or prevent thrips (Thysanoptera), fruit worms (Byturidae), and leaf diseases. Weeds were removed by hand.

### 2.4 Cover crops and tomato biomass, carbon and nitrogen contents

Aboveground fresh weight (FW) biomass yield of cover crops was determined using all the plant residues obtained in a plot (2.5 m\(^2\)) in both years. Cover crops samples were taken and dried in an oven at 60°C to determine their dry matter content which was used as a conversion factor to calculate the biomass yield. In October 2017, after the last tomato harvest, four tomato plants were taken from each replicate by cutting them at grafting point for measuring FW. In 2018, tomato shoot biomass yield was determined using all test plants. In both years, after the measurement of FW, three
The samples for MBN (moist bulk soil) were collected from the 0- to 10-cm depth at 4 and 8 WAT in 2017. In 2018, soil samples were collected from the 0- to 10-cm at 2 and 8 WAT (bulk soil), and also following harvest (bulk soil and rhizosphere soil, the latter of which was considered the soil attached to the roots). Soil samples were collected and immediately stored in a cool chamber at 4°C. Microbial biomass N was determined by a chloroform–fumigation–extraction method (Brookes, Landman, Prudent, & Jenkinson, 1985) followed by persulfate (50 g K₂S₂O₈, 16.8 g NaOH, 30 g H₃BO₃ L⁻¹) digestion (Doyle, Weintraub, & Schimel, 2004). Briefly, a 2- to 4-g sample was weighed in 30-ml glass vials and exposed to alcohol-free chloroform (CHCl₃) vapor in a vacuum desiccator for 24 h in dark at 25°C. After removing the CHCl₃, N was extracted from fumigated and nonfumigated samples with 10–20 ml of 0.5 M K₂SO₄ for 30 min on an oscillating shaker. Two ml of 0.5 M K₂SO₄ extract was mixed with 2 ml persulfate and then samples were stored for 2 d at room temperature before heating at 80°C overnight in a drying oven (DS-64, Yamato Scientific, Tokyo, Japan). Digested samples were used to analyze N as NO₃⁻—N—because persulfate oxidation converts dissolved organic N and NH₄⁺ to NO₃⁻—with a flow injection analyzer (SNAP-5000; Aqualab, Tokyo, Japan). The difference between NO₃⁻—N extracted from fumigated and nonfumigated samples (E fantasies) was converted into MBN using a conversion factor KEN with a value of 0.54, such that MBN = Eₙ/0.54 (Brookes et al., 1985).

### 2.5 Soil organic carbon and nitrogen pool

Soil samples in 2017 were collected from the 0- to 10-cm depth at 4 and 8 wk after transplanting (WAT). Following harvest in 2017, samples were collected from the 0- to 10- and 50- to 60-cm soil depths. In 2018, soil samples were collected from the 0 to 10 cm depth at 2 and 8 WAT. Following harvest in 2018, soil samples were collected from the 0- to 10-, 10- to 20-, and 20- to 30-cm depths. In both years, soil samples were composited within a depth for each treatment, dried at room temperature, sieved with 2-mm mesh, and then stored in a cool chamber at 4°C until analysis of C and N contents by combustion at 950°C with an elemental analyzer (Vario EL III; Elementar, Hanau, Germany). Because pH in soil samples was less than 7.0, soil total C was considered SOC (Nelson & Sommers, 1996). To determine bulk density, intact soil cores were collected following harvest each year from the 0- to 10- and 10- to 30-cm depths by a hand-pressing sampler that contained a steel volumetric ring (100 cm³). Three core samples were taken from each plot and then transferred to a paper bag and dried in an oven at 105°C until constant weight. Because bulk density at a soil depth did not vary significantly with treatment each year, averaged bulk density values across treatments were used each year to calculate SOC and STN (Mg ha⁻¹).

The Bulk density values at the 0- to 10- and 10- to 30-cm depths were 0.96 and 1.17 Mg m⁻³ in 2017 (Site 1), and 1.02 and 1.16 Mg m⁻³ in 2018 (Site 2), respectively. Soil organic carbon and STN were calculated using the following equation:

\[
\text{Soil C or N (Mg ha}^{-1}\text{)} = (Y / 100)(10^4 \text{m}^2\text{ha}^{-1})D_b d
\]

where \(Y\) is soil C or N content, \(D_b\) is bulk density (measured in Mg m⁻³), and \(d\) is soil thickness (measured in m).

### 2.6 Tomato yield, and nitrogen uptake and recovery

To determine tomato yield, red-ripe fruit was collected from six plots in each plot from 17 July to 25 September 2017, and from 3 August to 9 October 2018. Harvested fruit was classified in two categories: marketable, FW ≥100 g with no physiological disorders; and unmarketable fruit, FW <100 g and/or with physiological disorders such as misshapen fruit, blossom-end-rot, black spots, and huge scars (unmarketable yield data are not shown). Tomato yield (Mg ha⁻¹) was calculated by multiplying the total fruit weight of one plant by the plant density. Nitrogen uptake (Nup) was calculated as follows:

\[
\text{Nup (kg ha}^{-1}\text{)} = aY + bZ
\]

where \(a\) is N content of the shoot (average of N contents of stem and leaves), and \(Y\) is the total dry weight (DW) biomass of the shoot in each treatment; \(b\) is the N content of tomato fruit, and \(Z\) is the total DW of fruit (marketable and unmarketable) in each treatment. Nitrogen recovered from cover crops or slow-release N fertilizer (LPS100) was...
Table 2. Cover crops biomass yield, and carbon and nitrogen inputs in the soil in 2017 and 2018. Hairy vetch ‘Kantaro’, a winter-hardy cultivar (planted in fall, before snowfall), was used in 2017; whereas hairy vetch ‘Mamesuke’, a non-winter-hardy cultivar (planted in spring), was used in 2018.

| Treatments | Hairy vetch | Rye | Total N applied | Total C applied | C/N |
|------------|-------------|-----|----------------|----------------|-----|
|            | DW kg ha⁻¹ | N content g kg⁻¹ | N applied kg ha⁻¹ | DW | N content g kg⁻¹ | N applied kg ha⁻¹ |        |
| HVI        | 6,803       | 44.4 | 302             | –   | –               | –               | 302a¹   |
|            | 3,028b      | 10.0 |                  |      |                 |                 |         |
| HVM        | 6,756       | 44.4 | 300             | –   | –               | –               | 300a¹   |
|            | 3,007b      | 10.0 |                  |      |                 |                 |         |
| HV+RYE     | 3,414       | 44.4 | 152             | 4,608| 9.91           | 45.7            | 3,465a¹ |
|            | 2,639a      | 61.3 |                  |      |                 |                 |         |
| 2018       | HVI         | 2,489| 39.9            | 99.3| –               | –               | 99.3b   |
|            | HVM         | 2,427| 39.9            | 96.8| –               | –               | 96.8b   |
|            | RYE         | –    | –               | 6,189| 6.96           | 43.1           | 2,639a¹ |
|            | HV+RYE      | 1,502| 39.9            | 59.9| 5,048          | 11.3           | 2,777a  |
|            |             |      |                 |      |                 |                 | 61.3    |

¹HVI, hairy vetch incorporation; HVM, hairy vetch mulch; HV+RYE, biculture of hairy vetch and rye; RYE, rye monoculture; DW, dry weight.
²Means followed by the same letters in each column and year are not significantly different at 5% by Tukey’s honestly significant difference test.

Calculated with Equation 1 or Equation 2. It’s assumed that soil N uptake and/or fertilizer N uptake by tomatoes was similar in all treatments each year.

\[
\text{N recovery}_{\text{cover crops}} \% = \frac{\text{N}_{\text{up}}(\text{HVI}, \text{HVM}, \text{RYE, HV+RYE}) - \text{N}_{\text{up}}(\text{BARE-F})}{\text{cover crop N input}} \times 100 (1)
\]

\[
\text{N recovery}_{\text{fertilizer}} \% = \frac{\text{N}_{\text{up}}(\text{BARE-F}) - \text{N}_{\text{up}}(\text{BARE-NF})}{\text{fertilizer N input}} \times 100 (2)
\]

Where BARE-NF is no cover crop and no N fertilization.

2.7 Statistical analysis

The significance of mean differences among treatments each year was tested using analysis of variance (ANOVA) and Tukey’s honestly significant difference (Tukey’s HSD) tests in R software version 3.5.3 (R Core Team, 2019). For evaluating the significance of differences between two mean values, the t-test was used. Differences were accepted as significant if \( P \leq 0.05 \), unless otherwise stated.

3 RESULTS AND DISCUSSION

3.1 Cover crops biomass yield, and carbon and nitrogen inputs

Cover crops biomass yield, and C and N inputs varied (\( P < 0.05 \)) with treatments in each year (Table 2). The HV ‘Kantaro’ (overwintering type) and rye grew vigorously after winter in 2017. The total biomass yield and C input were 1.2-fold greater in HV+RYE than in HVI and HVM. However, because of the increased N content of HV residues than that of rye residues, N input was 1.5-fold greater in HVI and HVM than in HV+RYE.

In 2018, HV+RYE exhibited 2.7-fold greater biomass, and 2.8-fold greater C input than those in HVI and HVM (Table 2). RYE also exhibited greater biomass and C input than those in HVI and HVM. The increased biomass yield and C input in the biculture compared to those in the HV monoculture were the result of a greater planting density because the seeding rate used in the biculture was greater than that used in the monoculture. As opposed to 2017, N input was 1.2-fold greater in the biculture plots than in HV incorporation and mulch plots in 2018 because of an increase in N content of rye residues, in addition to least HV monoculture biomass yield. Greater N content of rye residues in the biculture than in monoculture was the result of N transfer from HV (Sainju et al., 2005).

The availability of cover crop N to subsequent crop is largely dependent on the C/N ratio of residues (Clark et al., 1994). Only rye monoculture showed a C/N ratio >25, capable of N immobilization and slow N release (Allison, 1966). The least HV biomass production in 2018 contributed greatly to the rise of the C/N ratio of the biculture.

3.2 Tomato yield, and nitrogen uptake and recovery

Tomato fruit yield and N uptake varied (\( P < 0.05 \)) with treatments in 2017 and 2018 (Table 3). In 2017, the marketable and total yields were 11.1–32.7% greater in cover crop treatments, especially HV+RYE, than in the BARE−F treatment.
effects of cover crop management and nitrogen fertilization on microbial biomass nitrogen in 2018

| Treatments | Yield | Nitrogen |
|------------|-------|----------|
|            | Marketable Mg ha⁻¹ | Total Mg ha⁻¹ | Uptake kg ha⁻¹ | Recovery % |
| 2017       |       |          |               |             |
| BARE–F     | 101c  | 117c     | 186c          | –           |
| HVI        | 116b  | 139ab    | 313a          | 42.0        |
| HVM        | 115b  | 130b     | 267b          | 27.0        |
| HV+RYE     | 134a  | 147a     | 312a          | 63.9        |
| 2018       |       |          |               |             |
| BARE–NF    | 51.6c | 73.8d    | 142c          | –           |
| BARE–F     | 60.1b | 86.2c    | 184b          | 28.0        |
| HVI        | 84.3a | 124a     | 233a          | 50.7        |
| HVM        | 64.8b | 102b     | 195b          | 11.4        |
| RYE        | 49.8c | 81.4cd   | 153c          | –71.8       |
| HV+RYE     | 60.7b | 91.8bc   | 188b          | 3.42        |

*Means followed by the same letters in each column and year are not significantly different at 5% by Tukey’s honestly significant difference test.

Tomato N uptake was 43.5, 67.7, and 68.3% greater in HVM, HV+RYE, and HVI, respectively, than in BARE–F. In 2018, HVI showed the greatest marketable and total yields (Table 3). Hairy vetch mulch and HV+RYE showed similar marketable yield to BARE–F, but the total yield was 6.5 and 18.3% greater in HV+RYE and HVM, respectively, than in BARE–F. Tomato N uptake was 2.17, 5.98, and 26.3% greater in HV+RYE, HVM, and HVI, respectively, than in BARE–F. However, RYE and BARE–NF showed the lowest marketable and total yields, and N uptake.

Greater fruit yield and N uptake in HVI, HVM, and HV+RYE than in BARE–F in 2017 and 2018 (Table 3) may be the result of more soil N availability in cover crops, especially with HV because of high N content and low C/N ratio. For similar reasons, fruit yield and N uptake were greater in BARE–F than in BARE–NF in 2018. The positive influence of HV mulch on tomato yield was reported by other researchers (Abdul-Baki, Teasdale, & Korcak, 1997; Abdul-Baki, Teasdale, Korcak, Chitwood, & Huettel, 1996; Araki, Hane, Hoshino, & Hirata, 2009). Our results suggest that the incorporation of HV residues may be more effective than HVM in increasing tomato yield.

The fact that N uptake and N recovery were greater in HVI than in HVM in both years (Table 3), despite a similar N input (Table 2), suggests that N release from HV residues was influenced by residue management. Generally, the decomposition rate of surface-placed residues (mulch) is lower than that of incorporated residues because the surface-placed residues have smaller soil–residue contact and residue–surface susceptible to microbial attack (Coppens et al., 2007; Summerell & Burgess, 1989). Also, surface-placed residues are subjected to alternating dry and wet periods and fluctuating temperature that may restrict microbial activity.

Greater tomato yield and N recovery in HV+RYE than in HVI and HVM in 2017, despite a lower N input in HV+RYE, may be the result of better synchrony of N release from biculture residues and tomato N demand, possibly because of a moderate decomposition speed of residues compared with HV incorporation (fast decomposition speed) and HV mulch (slow decomposition speed). As the C/N ratio of residues increases, N immobilization potential also increases and N release rate slows (Dabney, Delagado, & Reeves, 2001). Because of the increase in C/N ratio of residues from 17.6 in 2017 to 23.7 in 2018, HV+RYE showed the fewest effects on tomato yield and N recovery in 2018. The lowest tomato yield or negative recovery in RYE treatment would be expected because of C/N ratio > 25 (Table 2; Allison, 1966).

### 3.3 Microbial biomass nitrogen

The MBN levels varied ($P < .05$) with treatments in 2017 and 2018. In 2017, MBN levels were 25.4–175% greater in cover crop treatments, especially HVI and HV+RYE, than in BARE–F at 4 and 8 WAT (Figure 2).

In 2018, cover crop treatments exhibited 40.4–110% greater MBN levels than BARE–F at 2 WAT (Table 4). Likewise, cover crops treatments, except HVM, exhibited 91.1–186% greater MBN levels than BARE–F at 8 WAT. HV+RYE and HVI were the most effective treatments at 2 and 8 WAT, respectively. However, cover crop management effects on
MBN varied \( (P < .05) \) with sampling sites following harvest (17 WAT; Table 4). The HVI and HV+RYE treatments showed the lowest MBN levels in the bulk soil. However, HV showed the highest MBN levels in the rhizosphere soil, while RYE and HV+RYE showed the lowest MBN levels. Averaged across treatments, MBN levels were 37.3% higher in rhizosphere than in bulk soil, possibly because rhizodeposition stimulates microbial growth (Nannipieri et al., 2008).

Regardless of the C/N ratio or residue placement, above or below ground, all plant residues that are returned to the soil enhance MBN levels during the early decomposition stages, because if plant residue N does not satisfy the microbial growth requirements, microbes use inorganic N from the soil (Bird, Horwath, Eagle, & van Kessel, 2001; Chen et al., 2014). This may explain the increase in MBN levels in all cover crop treatments. Overall, the incorporation of HVI was the most effective cover crop treatment in increasing MBN levels.

Microbial growth becomes limited following depletion of mineral nutrients in the rhizosphere, and nitrogen is one of the major nutrients required by soil microbes for their propagation. Therefore, the application of plant residues of high C/N ratio or low N content to the soil may result in limited microbial growth in the rhizosphere due to low N availability from plant residues and the soil (rhizosphere position increases soil C/N ratio; Chen et al., 2014; Marschner, 1995; Nannipieri et al., 2008). In this sense, the lowest MBN levels in the rhizosphere soil in RYE and HV+RYE in 2018 (Table 4) may have resulted from low soil N availability due to high C/N ratio of residues applied to the soil (the C/N ratio of RYE and HV+RYE were 23.7 and 61.3, respectively, 2.32- to 6.01-fold greater than that of HVI and HVM; Table 2). The lowest MBN levels at rhizosphere soil in RYE and HV+RYE were consistent with their lowest N recovery values. This fact suggests that MBN levels at rhizosphere soil may be more important than MBN at bulk soil in predicting tomato N uptake from plant residues.

Microbial biomass N controls soil inorganic N availability and loss, especially in high input systems (Moore, Klose, & Tabatabai, 2000). Therefore, the increase in MBN in cover crop treatments may have contributed to an increase in soil N availability, especially in the early period after residue incorporation (2–4 WAT) in both years (Figure 3). Averaged across sampling dates, in 2017, soil N availability increased by 1.26, 10.2, and 33.7% in HVM, HV+RYE, and HV, respectively; while in 2018, soil N availability increased by 9.02, 38.4, and 56.6% in HV+RYE, HVM, and HV, respectively, compared with BARE−F. In contrast, soil N availability decreased by 4.32% in RYE compared with BARE−F, possibly as the result of microbial N immobilization. Increased MBN levels with N fertilization compared to no N fertilization in 2018 (Table 4) was possibly the result of microbial N immobilization (Balabane & Balesdent, 1992; Mulvaney, Khan, & Ellsworth, 2009; Shen, Hart, Powlson, & Jenkinson, 1989).

### 3.4 Soil residual inorganic nitrogen

The NO\(_3^-\)−N and NH\(_4^+\)−N levels in the soil following tomato harvest varied \( (P < .05) \) with treatments at 0- to 10-cm depth in 2017 (Table 5). Hairy vetch mulch showed the greatest NO\(_3^-\)−N and NH\(_4^+\)−N levels. Hairy vetch incorporation showed greater NO\(_3^-\)−N levels than HV+RYE and BARE−F, whereas NH\(_4^+\)−N levels in HVI were similar to those in HV+RYE and BARE−F. As the result of increased NO\(_3^-\)−N and NH\(_4^+\)−N levels, soil inorganic N (SIN) levels were 25.1 and 96.9% greater in HVI and HVM, respectively, than in BARE−F. As opposed to 0- to 10-cm depth, the effects of cover crops were only significant \( (P < .05) \) for NH\(_4^+\)−N at 50- to 60-cm depth (Table 5). The HVM, HVI, and HV+RYE treatments, especially HVI, showed greater NH\(_4^+\)−N than the BARE−F treatment. Similarly, SIN levels increased in HVI (60.7%) more than in HVM (14.2%) and HV+RYE (10.7%), compared with BARE−F.
Increased NO\textsubscript{3}\textsuperscript{−}−N and NH\textsubscript{4}\textsuperscript{+}−N levels at 0- to 10-cm soil depth in HVM than in HVI may have resulted from lower tomato N uptake (Table 3) and delayed N release from mulch residues because of slow decomposition (Bradford & Peterson, 2000; Coppens et al., 2007). On the other hand, lower NO\textsubscript{3}\textsuperscript{−}−N or SIN levels at 0- to 10-cm in HV+RYE than in HVM may have resulted from higher N uptake in HV+RYE. Greater NH\textsubscript{4}\textsuperscript{+}−N or SIN levels at 50- to 60-cm depth in cover crop treatments than in the bare treatment may be the result of decomposition of cover crop roots. The fact that HVI showed remarkably greater NH\textsubscript{4}\textsuperscript{+}−N or SIN levels at 50- to 60-cm depth than HVM and HV+RYE (Table 5) suggests that N may have leached from the topsoil to deeper soil layers in HVI plots, possibly due to the fast decomposition of HV residues after incorporation in the soil. However, because of low mobility of NH\textsubscript{4}\textsuperscript{+} in soils,—NH\textsubscript{4}\textsuperscript{+} is retained by soils that are negatively charged in the temperate region (Di & Cameron, 2002)—N may have leached mostly as dissolved organic N that can further be converted into NH\textsubscript{4}\textsuperscript{+} in deeper soil layers (Murphy et al., 2000; Van Kessel, Clough, & van Groenigen, 2009).

Cover crops and N fertilization influenced ($P < .05$) the NO\textsubscript{3}\textsuperscript{−}−N and NH\textsubscript{4}\textsuperscript{+}−N levels at 0- to 30-cm depth in 2018 (Table 6). Cover crop treatments showed greater NO\textsubscript{3}\textsuperscript{−}−N levels than the bare treatments at all depths, especially at 0- to 10-cm depth. Nitrogen fertilization (BARE−F) increased NO\textsubscript{3}\textsuperscript{−}−N at 0- to 10-cm depth only compared with no N fertilization (BARE−NF). As for NH\textsubscript{4}\textsuperscript{+}−N, cover crop treatments increased NH\textsubscript{4}\textsuperscript{+}−N levels at 0- to 10- and 10- to 20-cm depths, except HVM at 10- to 20-cm depth, compared with bare treatments. However, at 20- to 30-cm depth, cover crop treatments tended to show similar NH\textsubscript{4}\textsuperscript{+}−N levels to the bare treatments. As a result of increased NO\textsubscript{3}\textsuperscript{−}−N and NH\textsubscript{4}\textsuperscript{+}−N levels, SIN levels were 12.2–218% greater in cover crop treatments than in BARE−F at 0- to 30-cm depth (Table 6).

As in 2017, HVM showed greater residual nitrate-N or inorganic N at 0- to 10-cm than any other cover crop treatment in 2018 because of delayed N release from mulch residues.
(Bradford & Peterson, 2000). Likewise, greater residual inorganic N at 0- to 10-cm in RYE than in HVI or HV+RYE may be explained by least tomato N uptake in RYE and delayed N release from rye residues due to high C/N ratio. Overall, the results from 2017 and 2018 suggest that in northern Japan, under high tunnel conditions, N leaching potential may be higher under HV monoculture, especially when residues are placed on the soil surface (mulch), than under the biculture of HV and rye. Due to the fast decomposition of residues, HV incorporation may show increased N leaching potential in the early period after residue incorporation, while HV mulch, with a slow decomposition speed, may show increased N leaching potential in the late period. Moreover, the results from 2017 suggest that if adequate seeding HV to rye ratio is used, residual N following biculture management may be negligible because of increased residue-derived N recovery by tomatoes. Therefore, the use of biculture may represent a better opportunity to increase crop yields with the least negative effect on the environment.

### 3.5 Soil organic carbon and total nitrogen

The SOC and STN at each depth varied ($P < .05$ or $P < .1$) with treatments in 2017 and 2018 (Table 7). In 2017, SOC increased by 2.87, 5.94, and 8.81% in HV+RYE, HVI, and HVM, respectively, compared with BARE–F at 0- to 10-cm depth. However, at 10- to 30-cm depth, SOC increased in HVI (7.08%) and HV+RYE (2.36%) only, compared with

### Table 5 Effects of cover crop management on soil inorganic nitrogen ($\text{NO}_3^-\text{N} + \text{NH}_4^+\text{N}$) at 0- to 10- and 50- to 60-cm depths after tomato harvest in 2017

| Treatments | SOC$^c$ $\text{Mg C ha}^{-1}$ | STN $\text{Mg N ha}^{-1}$ | MBN/STN ratio |
|------------|-------------------------------|---------------------------|---------------|
|            | 0–10 cm | 10–30 cm | Mean  | 0–10 cm | 10–30 cm | Mean |
| 2017       |         |          |       |         |          |       |
| BARE–F     | 52.2c   | 127b     | 89.5c | 3.10b   | 7.37bc   | 5.23b |
| HVI        | 55.3ab  | 136a     | 95.5a | 3.45a   | 7.97a    | 5.71a |
| HVM        | 56.8a   | 127b     | 91.9ab| 3.20b   | 7.25c    | 5.23b |
| HV+RYE     | 53.7bc  | 130ab    | 91.8bc| 3.22b   | 7.50b    | 5.36b |
|            |         |          |       |         |          |       |
| 2018       |         |          |       |         |          |       |
| BARE–NF    | 29.2b   | 58.3b    | 43.8c | 2.39b   | 5.53b    | 3.96c |
| BARE–F     | 29.0b   | 63.0a    | 46.0b | 2.47b   | 5.94a    | 4.21b |
| HVI        | 34.3a   | 63.7a    | 49.0a | 2.78a   | 5.99a    | 4.39a |
| HVM        | 34.0a   | 63.4a    | 48.7a | 2.73a   | 5.92a    | 4.32a |
| RYE        | 34.4a   | 62.4ab   | 48.4a | 2.75a   | 5.92a    | 4.33a |
| HV+RYE     | 34.5a   | 62.1ab   | 48.3a | 2.72a   | 5.80ab   | 4.26ab |

aSOC, soil organic carbon; STN, soil total nitrogen; MBN, microbial biomass nitrogen; BARE–F, no cover crop with nitrogen fertilization; HVI, hairy vetch incorporation; HVM, hairy vetch mulch; HV+RYE, biculture of hairy vetch and rye; BARE–NF, no cover crop without nitrogen fertilization; RYE, rye monoculture.

bMeans followed by the same letters in each column are not significantly different at 5% by Tukey’s honestly significant difference test.

cna, not applicable.

**Table 6 Effects of cover crop management and nitrogen fertilization on soil inorganic nitrogen ($\text{NO}_3^-\text{N} + \text{NH}_4^+\text{N}$) at 0- to 30-cm depth after tomato harvest in 2018**

| Treatments | $\text{NO}_3^-\text{N} + \text{NH}_4^+\text{N}$ $\text{mg N kg}^{-1}$ | $\text{NO}_3^-\text{N} + \text{NH}_4^+\text{N}$ $\text{mg N kg}^{-1}$ | $\text{NO}_3^-\text{N} + \text{NH}_4^+\text{N}$ $\text{mg N kg}^{-1}$ |
|------------|---------------------------------------------------------------|---------------------------------------------------------------|---------------------------------------------------------------|
|            | 0–10 cm                                                      | 50–60 cm                                                      | |
| BARE–F     | 4.77c                                                        | 23.9b                                                         | 28.7c                                                         |
| HVI        | 9.50b                                                        | 26.4b                                                         | 35.9b                                                         |
| HVM        | 23.2a                                                        | 33.3a                                                         | 56.5a                                                         |
| HV+RYE     | 4.27c                                                        | 23.7b                                                         | 28.0c                                                         |

aBARE–F, no cover crop with nitrogen fertilization; HVI, hairy vetch incorporation; HVM, hairy vetch mulch; HV+RYE, biculture of hairy vetch and rye.

bMeans followed by the same letters in each column are not significantly different at 5% by Tukey’s honestly significant difference test.

Greater SOC in cover crop treatments than in bare treatments at 0- to 10-cm depth in 2017 and 2018 may have resulted from higher C input in the soil (Campbell, Selles, Lafond, & Zentner, 2001). Sainju et al. (2003) reported...
greater SOC at surface soil layer with HV and rye than with bare fallow (with or without N fertilization). As with SOC, the greater STN with cover crops than with no cover crops observed mostly in 2018 may be the result of higher N input by cover crop residues (Table 2), especially HV, due to its high N content. Our results agreed with those of Sainju et al. (2000), who reported increased soil organic N with HV, rye, and crimson clover (Trifolium incarnatum L.), especially HV, compared to the bare fallow (with or without N fertilization).

As opposed to HV+RYE and HVM, the HVI treatment was effective in increasing SOC at 0- to 30-cm depth and STN at 0- to 10-cm depth in both years (Table 7), compared with BARE−F. Therefore, incorporation of HV residues may be a better management practice in improving SOC and STN at surface soil layer in plastic high tunnel systems in short-term. This increased effectiveness may be related to the accelerated decomposition of HV residues (monoculture) when incorporated compared with when mulched or mixed with rye residues. However, over a long-term, the biculture of HV and rye may be more effective in building up SOC and STN than HV monoculture because of greater biomass production (Campbell et al., 2001; Kuo, Sainju, & Jellum, 1997b). This assumption is sustained by the results of Sainju, Singh, & Singh (2015), who reported greater soil C and N with the biculture of HV and rye than HV and rye alone or the control (no cover crop) under forage and sweet sorghum in a 4-year field examination.

### 4 CONCLUSIONS

Cover crop management influenced differently soil C and N pool, and tomato yield. Regardless of management, HV and rye treatments improved SOC at surface 10-cm soil depth. However, the positive effects of cover crops on STN varied with soil depths and years, and HV incorporation treatment tended to be more effective than other treatments in increasing STN. Residual soil nitrate−N, subject to leaching losses after winter, increased with HV monoculture more, especially when HV residues were placed on soil surface, than with the biculture of HV and rye. Microbial biomass N, soil N availability, and tomato yield increased in response to HV application, especially when residues were incorporated in the soil. However, the effects of the biculture of HV and rye on MBN, soil N availability, and tomato yield varied with C/N ratio of residues, and the best results were obtained with residues with a C/N ratio of 17.6 (obtained with a seeding HV to rye ratio of 2:1) than with 23.7. These findings partially supported our hypotheses. The results suggest that if adequate seeding HV to rye ratio is used, the biculture of HV and rye is a better management practice to increase SOC and STN at surface layer, and tomato yield with least soil residual N. However, because the effectiveness of the biculture is strongly determined by the C/N ratio of residues, in addition to the short period of this study, a study longer than two years is needed to clearly understand the benefits of the biculture on soil and environmental quality and tomato yield in northern Japan.

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### ORCID

Rafael A. Muchanga

https://orcid.org/0000-0002-7831-7409

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**TABLE 7** Effects of cover crop management and nitrogen fertilization on soil organic carbon (SOC) and total nitrogen (STN) at 0- to 30-cm depth in 2017 and 2018

| Treatments | 0−10 cm | 10−20 cm | 20−30 cm |
|------------|---------|----------|----------|
|            | NO$_3^-$−N | NH$_4^+$−N | SIN | NO$_3^-$−N | NH$_4^+$−N | SIN | NO$_3^-$−N | NH$_4^+$−N | SIN |
| BARE−NF    | 6.90bc    | 7.97bc   | 14.9e   | 2.23c    | 5.50c    | 7.73d   | 6.93c    | 8.27ab   | 15.2d |
| BARE−F     | 21.6d    | 5.90c    | 27.5d   | 5.13c    | 6.37c    | 11.5c   | 7.23c    | 8.43ab   | 15.6cd |
| HVI        | 37.1c    | 11.9a    | 49.0c   | 16.9a    | 7.67ab   | 24.6a   | 25.9a    | 7.37b    | 33.3a |
| HVM        | 76.8a    | 10.6ab   | 87.4a   | 9.90b    | 6.47c    | 16.4b   | 11.6b    | 8.63ab   | 20.2bc |
| RYE        | 49.4b    | 13.2a    | 62.6b   | 18.4a    | 7.83a    | 26.2a   | 9.00bc   | 8.47ab   | 17.5c |
| HV+RYE     | 33.3c    | 10.4ab   | 43.7c   | 11.0b    | 6.50bc   | 17.5b   | 14.8b    | 9.53a    | 24.3b |

aSIN, soil inorganic nitrogen; BARE-NF, no cover crop without nitrogen fertilizer; BARE-F, no cover crop with nitrogen fertilization; HVI, hairy vetch incorporation; HVM, hairy vetch mulch; RYE, rye monoculture; HV+RYE, biculture of hairy vetch and rye.

*Means followed by the same letters in each column and year are not significantly different at 5% or 10% by Tukey’s honestly significant difference test.*
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