

Towards the development of cover crop - reduced tillage systems without herbicides and synthetic fertilizers in onion cultivation: Promising but challenges remain

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ABSTRACT

Cover crops with reduced tillage technology (CC-RT) can foster soil health and functioning, a crucial agroecological principle in any transition strategy to more sustainable agricultural systems. However, CC-RT commonly strongly relies on herbicides and synthetic fertilizers, and vegetable crop yields are variable and often low. We assessed the effects of two tillage systems (RT and conventional tillage) and the application of native effective microorganisms (NEM) on onion crop growth and development, yield, N-status, weed pressure, and soil physico-chemical and biological quality after a summer CC, without using herbicides or synthetic fertilizers. Using a participatory research strategy, we conducted a two-year experiment at an experimental station and a one-year trial on two commercial farms. Onion yields were generally low (between 10 and 16 Mg ha⁻¹) and lower in 2019 than in 2020, and lower in RT than in CT in 2020. The relatively low yields in 2019 and RT were associated with poor crop growth and development and leaf-N concentrations below the critical threshold in the early stages of crop development. Soil bulk density was not limiting crop growth in any treatment. Soil mineral N was lower in 2019 than in 2020 and did not significantly differ between treatments. Soil biological activity was higher in RT than in CT. Although the CC residue soil cover in the early stages of the onion crop in RT was more than 50%, RT had a higher weed pressure than CT, which was reversed later in the growing season. The NEM application did not significantly affect most crop, weed, and soil variables. In conclusion, a reduced tillage system for onions without herbicides and synthetic fertilizers is within reach. However, further research is needed to manage weeds and soil N supply dynamics to make CC-RT feasible for no or low agrochemical input systems.

1. Introduction

Maintaining soil health is fundamental for reaching global sustainability goals (Kraamwinkler et al., 2021; Lehmann et al., 2020) and is a crucial agroecological principle in any transition strategy to more sustainable agricultural systems (Hoffland et al., 2020; Nicholls et al., 2016; Wezel et al., 2020). Soil health can be fostered by using wide crop rotations, limiting the proportion of root and tuber crops, using pasture, cover crops, green manure and organic amendments (Alliaume et al.,

2013; Dogliotti et al., 2014; King and Blesh, 2018), reducing pesticide use (Hussain et al., 2009; Rose et al., 2016), and implementing erosion control practices (Alliaume et al., 2013; Dogliotti et al., 2014). However, these practices to maintain or improve soil health may not always be feasible. For example, maintaining soil organic carbon (SOC) levels in vegetable systems on soils with a high SOC level may require unrealistically high input levels of organic matter (Alliaume et al., 2013). In addition, three to four years of pastures do not always fit in small vegetable-based crop rotation systems, and therefore cash-crop

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dominated rotations are often prone to erosion (García De Souza et al., 2011). Therefore, a broader set of management practices is needed to maintain soil health in vegetable systems.

The use of cover crops (CC) combined with reduced tillage (RT) is a promising practice that can minimize soil erosion (Alliaume et al., 2014), increase and maintain high SOC levels (Haddaway et al., 2017; Li et al., 2020), increase water infiltration (Alliaume et al., 2014), enhance biological activity and biological control (Navarro-Miró et al., 2022; Tamburini et al., 2016), reduce N leaching (Jokela and Nair, 2016; Zhang et al., 2020) and reduce CO₂ emissions (Abdalla et al., 2013; Boeckx et al., 2011). However, CC-RT management often results in variable and low crop yields, and farmers face technical difficulties implementing this system (Alliaume et al., 2014; Erenstein, 2002; Jokela and Nair, 2016; Navarro-Miró et al., 2022, 2007; Peigné). Thus, while promising, CC-RT needs to be further developed to become a practically feasible management option.

The implementation of CC-RT will influence the crop system as a whole, affecting the topsoil temperature, water and soil organic matter dynamics, and nutrient supply (Alliaume et al., 2014; Cook et al., 2006; Jokela and Nair, 2016; Peigné et al., 2007; Tittarelli et al., 2018). Therefore, the CC-RT systems must be tailored to the site-specific conditions to bridge the yield gap compared to conventional tillage systems. Moreover, most CC-RT systems strongly rely on herbicides to terminate the CC and to control weeds and on synthetic N-fertilizers to ensure crop N-uptake at initial crop development stages (Antichi et al., 2022; Carr et al., 2013; Farooq and Siddique, 2015). Therefore, implementing CC-RT poses additional challenges in no or low agrochemical input systems (Casagrande et al., 2016; Peigné et al., 2007; Vollmer et al., 2010; Carr et al., 2013).

Organic agricultural systems rely on mechanical practices for weed control and CC termination. The effectiveness of CC-RT and its residues in controlling weeds is erratic, and reducing tillage intensity is seen as a bottleneck (Carr et al., 2013; Casagrande et al., 2016; Mandal et al., 2021; Peigné et al., 2015). Additionally, crop nutrition in organic systems mainly relies on organic matter management. Since the time and amount of N release from organic amendments and soil organic matter is difficult to predict (Geisseler et al., 2022; Hodge et al., 2000; Masunga et al., 2016), matching N supply and crop demand under organic CC-RT management is challenging. The decomposition of the CC residues may result in N immobilization, depending on the CC biomass and C:N ratio (Hodge et al., 2000; Mooshammer et al., 2014; Masunga et al., 2016). Early termination of the CC and incorporating legume species in the CC may lower the C:N ratio and, thus, the N-immobilization risk (Ranells and Waggener, 1996). Manipulation of the structure and functions of the microbiota through microbial inoculants could stimulate soil organic matter mineralization and promote nutrient availability for the crop (Terrazas et al., 2016). The application of effective microorganisms (Alarcon et al., 2020; Higa and Wididana, 1991; Morocho and Mora, 2019; Singh et al., 2011), particularly when combined with organic amendments (Khaliq et al., 2006), may promote this effect. However, strategies that reduce N immobilization risk or increase N mineralization may decrease soil cover and weed suppression, indicating a trade-off between soil cover and N supply.

Onion is one of the main vegetable crops in the world (FAO, 2021). Onion has a shallow, sparse, and low-density root system (Geisseler et al., 2022), responds strongly to nitrogen availability (Brewster and Butler, 1989; Geisseler et al., 2022), and is highly susceptible to weed competition (Hewson and Roberts, 1973; van Heemst, 1985). Therefore, onion production heavily rely on herbicides and synthetic fertilizers in conventional systems and on mechanical tillage and labour for weeding in organic systems. Low soil cover after removal of weeds makes onion cultivation prone to soil erosion and nutrient leaching. Developing CC-RT systems for onions without agrochemical inputs, particularly herbicides and synthetic fertilizers, could significantly reduce agrochemical use and soil erosion.

We conducted a two-year study to assess the effects of the tillage

system (reduced tillage vs conventional tillage) and the application of native effective microorganisms (presence vs absence) on onion production after a summer cover crop, without using herbicides or synthetic fertilizers. We conducted experiments at an experimental station field and at two commercial farms using a participatory research approach. This paper reports on onion crop growth and development, onion yield, N-status, and weed pressure as main response variables, as well as soil physical, chemical, and biological properties as supporting variables to comprehensively understand the crop system performance.

2. Materials and methods

2.1. Study area and research approach

The study was conducted in Canelones Department, South Uruguay, where most vegetable production of the country is concentrated (34°21'S to 34°57'S – 55°40'W to 56°40'W). The climate is humid subtropical, with an average mean temperature of 17 °C (average minimum: 11 °C, average maximum: 23 °C) and light frosts between May and September. Mean annual precipitation is 1200 mm, evenly distributed throughout the year, but with significant variation between years (Castaño et al., 2011). The main soil types are Mollic Vertisols (Hyper-eutric), Luvic/Vertic Phaeozems (Pachic), and Luvic Phaeozems (Abruptic/Oxyaquic) (Alliaume et al., 2013).

The study comprised a two-year experiment at the Centro Regional Sur Experimental Station (CRS) of the Faculty of Agronomy, Universidad de la República (see Section 2.3) and two simplified experiments on an organic and a conventional commercial farm in the second year (see Section 2.4, Appendix A). We used an interdisciplinary and participatory approach where a project support group of around 30 people, including farmers, technical advisers, and researchers, participated from the beginning of the study to define, monitor and assess, the operational and tactical management of the experiments. The project support group met in the first workshop in 2017, two field days and workshops each year in 2019 and 2020, and a final workshop in 2021. In addition, the farmers of the two commercial farms had weekly interactions with the research team.

2.2. Design criteria and analytical framework

During the starting workshop in December 2017, participants expressed their interest and discussed their difficulties implementing CC-RT. Onion production was highlighted as particularly challenging regarding CC and weed management without using herbicides, and nitrogen management without synthetic fertilizers. Therefore, onion was selected to study a CC-RT system without herbicides and synthetic fertilizer inputs, and the experiences and suggestions of the workshop participants to overcome challenges were used to inform the experimental treatments. The mutually agreed design criteria for effective CC-RT systems were: (i) high biomass production and fast soil cover development of the CC; (ii) CC species with low risk of becoming a weed; (iii) termination of the CC should be possible without herbicides; and (iv) the CC should have a C:N ratio below 30 to reduce the risk of nitrogen immobilization (Hodge et al., 2000; Mooshammer et al., 2014). Participants also expressed interest in the potential of local biological inputs, such as (native) effective micro-organisms (EM and NEM), to increase soil organic matter mineralization and crop nitrogen availability (Higa and Wididana, 1991; Olle and Williams, 2013).

We assessed key response variables related to the crop, the weeds, and the soil to capture the relationships between components of the CC-RT-onion cropping system (Fig. 1). Specifically, we assessed crop system outcomes in terms of crop growth and development, crop yield, crop N status, and weed pressure. We assessed soil chemical, physical, and biological properties, as well as CC and residue soil cover, to understand the underlying causes of these outcomes. This analytical framework guided the discussion at each workshop.

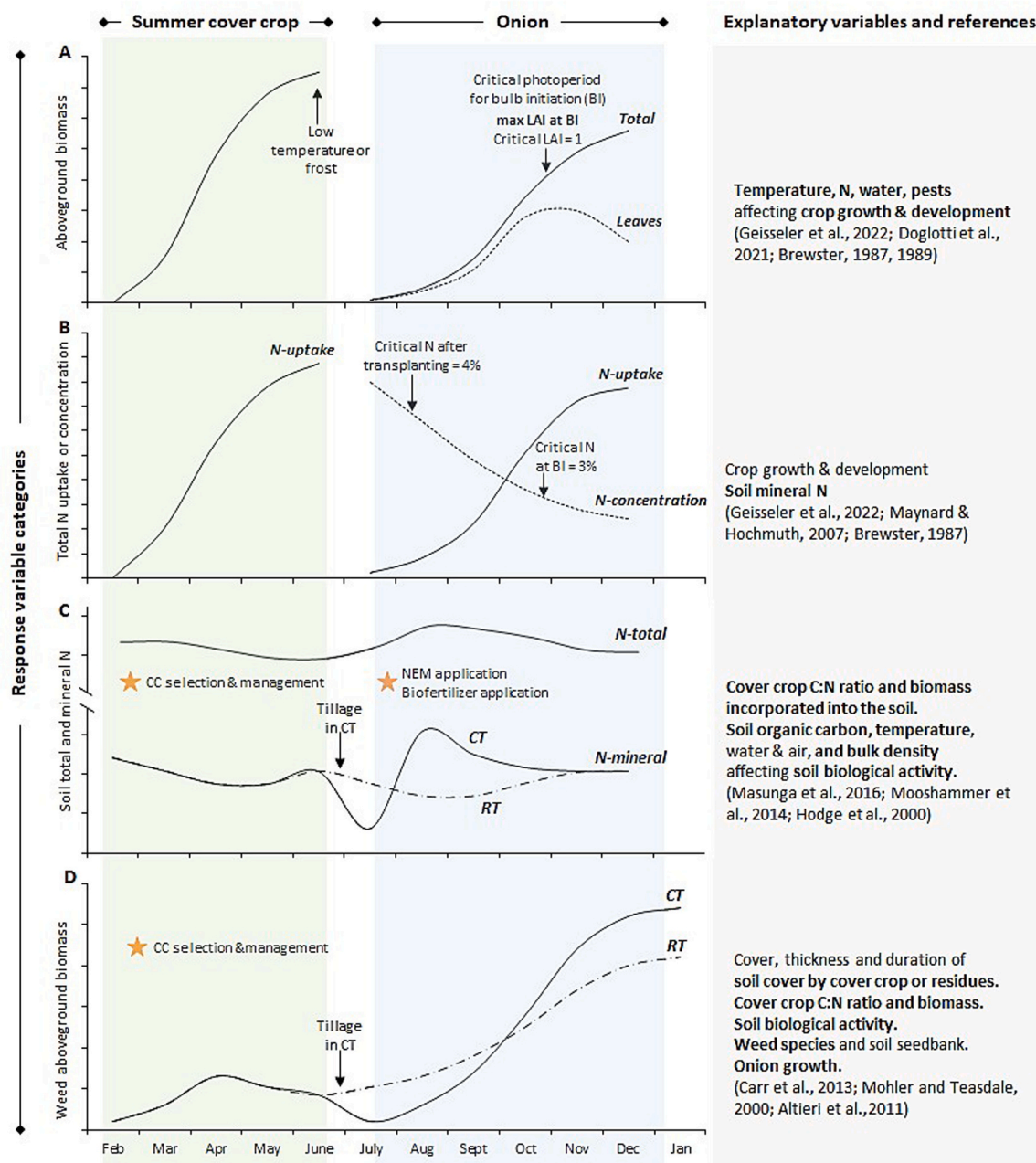


Fig. 1. Conceptual representation of the hypothesized trends along the year of the four response variable categories of the cover crop (CC) - onion cycle (left) and the explanatory variables that may explain the crop system performance (right). Main response variable categories: cover crop and onion aboveground biomass (A), crop nitrogen status (B), soil nitrogen (C), and weed pressure (D). Variables in bold letters correspond to the variables assessed. Orange stars correspond to the strategies to avoid N deficiencies and problematic weed pressure during the onion crop. BI is bulb initiation; solid lines indicate conventional tillage (CT); dashed lines indicate reduced tillage (RT).

2.3. CRS on-station experiment

2.3.1. Treatments and experimental design

The experiment was arranged in a randomized complete block design with four replications using a split-plot design. The whole plot factor was tillage (reduced (RT) and conventional (CT)), and the split-plot factor was NEM application (present+ or absent-), resulting in a total of four treatments (RT/NEM-, RT/NEM+, CT/NEM-, CT/NEM+). The experimental field was 1760 m², where each split-plot of 67.5 m² (4.5 × 15 m) consisted of three contiguous raised beds, which were 1.5 m apart and 15 m long (Appendix A). The experiment location and the experimental design were the same in both years, so each split-plot consistently received the same treatment in the two years.

2.3.2. Soil characterization

The soil at the experimental site was a Mollic Vertisol (Hypereutric) (USS Working Group WRB, 2006), with 10% sand, 42% silt and 48% clay in the upper soil layer. It had 2.55% SOC in both years, 1.8 and 1.7 cMol_c kg⁻¹ K, and 192 and 162 mg kg⁻¹ P-Bray 1 in 2019 and 2020, respectively. Bulk density was 0.89 and 0.84 g cm⁻³ in 2019 and 2020, respectively. No significant differences were detected in SOC, nutrient content, and bulk density between the split-plots before the application of treatments in 2019 and 2020 (p>0.1).

2.3.3. Soil and crop management

A sequence of a frost-sensitive summer CC consisting of foxtail millet (*Setaria italica*; sowing rate 30 kg ha⁻¹ in 2019 and 50 kg ha⁻¹ in 2020)

and cowpea (*Vigna unguiculata*; sowing rate 20 kg ha⁻¹) followed by an onion crop was established during two subsequent years (2019–2020, Appendix A). The CC was sown in raised beds at the end of the summer (22 February 2019, 11 March 2020) and finished its growing cycle due to low temperatures in the first half of June 2019 and at the end of May 2020 (Appendix D). Intermediate day-length onion varieties were used: Pantanoso del Sauce-CRS in 2019 and Armonía-CRS in 2020. Onion seedlings (aged 92 days) were transplanted by hand into raised beds on 6 and 4 August and harvested on 20 and 16 December in 2019 and 2020, respectively. Plant density was 214,000 plants ha⁻¹ arranged in three rows per bed (80 cm width), with a 20 cm distance between rows and from the edge of the raised-beds.

The experimental area was tilled annually during summer (January and February) before installing the CC. Soil management at the end of the CC and before transplanting onion in the CT treatment consisted of two chisel and disc ridger passes during June and July (25–30 cm), one rotavator with bed forming (20 cm), and a furrow opener (15 cm) before transplanting. The RT treatment consisted of mechanical crushing of the CC with an inverted tooth harrow, followed by one pass with a furrow opener (15 cm) before transplanting in 2019 and two passes with a furrow opener (15 cm) starting four days before transplanting in 2020 (detailed information in Appendix B). The NEM treatments consisted of immersion of the onion seedlings roots for two hours in a solution with 10% NEM before transplanting, plus ten soil applications of a NEM solution from transplanting in 2019 and ten soil applications from one week before transplanting to bulb initiation (BI) in 2020 (70 L ha⁻¹, dilution 10%). The NEM solution was obtained from a local company and was physico-chemically and microbiologically analyzed before use (Appendix C).

Before sowing the CC, chicken manure was applied and incorporated into the soil (11 and 14 Mg DM ha⁻¹ in 2019 and 2020, respectively, Appendix A), resulting in an estimated N supply from chicken manure of 112 and 168 kg N ha⁻¹ (Appendix B). An organic-N fertilizer (Mixamin, 7.2 g L⁻¹) was applied throughout the growing season of onion. In 2019, seven soil applications at a dose of 12.5 L ha⁻¹ and six foliar applications at 900 cc ha⁻¹ resulted in a total of 7 kg N ha⁻¹. In 2020, compost-tea (1.3 g L⁻¹) was also used: nine soil applications of organic-N fertilizer at a dose of 12.5 L ha⁻¹ and compost-tea at 8 L ha⁻¹ resulted in a total of 9 kg N ha⁻¹. Water was provided through drip irrigation at transplanting and during the onion growing season, based on daily visual monitoring of the field and the potential crop evapotranspiration (Allen, 2006). Pest and disease management for downy mildew caused by *Peronospora destructor* and *Sminthurus viridis* consisted of six fungicide applications (three copper sulphate, two copper+mancozeb+metalaxyl, one copper+mancozeb+cymoxanil) and one insecticide (azadirachtin) application in 2019, and five foliar applications of *Trichoderma spp.*, four fungicide (copper sulphate), and one insecticide (azadirachtin) application in 2020 (detailed information in Appendix B). Leaf-cutter ants were controlled with granular insecticide bait (Fipronil). Weed control at onion transplanting involved soil tillage in CT and manual weeding in RT. During the onion growing cycle, weeds were removed manually after weed pressure assessments. Three manual weeding operations were conducted in both tillage treatments in 2019, and three and four in CT and RT, respectively, in 2020.

2.3.4. Data collection

2.3.4.1. Cover crop biomass and quality, soil cover and weed pressure. The CC aboveground biomass and quality were assessed on 5 June 2019 and 25 May 2020, when plants started senescence due to low temperatures. The aboveground biomass was estimated by harvesting quadrants of 0.36 m² in three random replicates per plot. CC species and weeds were separated, dried at 60°C for 48 h, and weighted. The carbon content of the samples was assessed by oxidation with K₂CrO₇ in H₂SO₄ at 150°C for 30 minutes, followed by colourimetric determination (Mebius,

1960), and the N concentration was evaluated using the Kjeldahl method (Bremmer and Mulvaney, 1982). No significant differences in CC biomass and composition were detected between split-plots in either 2018 or 2019 (p>0.1).

The proportions of bare soil, CC cover, weed cover, and onion cover were estimated by sampling every 5 cm along a 1.6 m-transect using a PIN micro-relief meter (locally made following Alliaume et al., 2014) and recording the type of cover in three replicates per plot. CC residue cover was assessed on three 1-m transects on the soil surface per plot. Measurements were taken at sowing and the end of the CC, before onion transplanting, 30 and 60 days after transplanting, at BI, and before harvest in both years.

Weed aboveground biomass (g DM m⁻²) and dominant weed species were assessed in three randomly selected 0.36 m² squares per plot at the end of the CC, before transplanting, at 20 and 50 days after transplanting, at BI, and before onion harvest. Weeds were collected and dried at 60°C for 48 h.

2.3.4.2. Onion crop yield, growth and development, and foliar nitrogen.

Onion yield was measured in each plot by harvesting all the plants in 8 m of the central bed of a plot where no destructive sampling had taken place. Harvested onions were left to dry under sheltered, ambient conditions for one month. Then, leaves, false stems, and roots were removed, and total bulb yield, marketable yields (Mg ha⁻¹), number of bulbs, and average bulb size (g) were measured. Total yield comprised all bulbs, while marketable yield included bulbs greater than 4 cm in diameter.

Onion growth, development, and nitrogen status were measured by determining bulbing ratio, BI, aboveground biomass, number of leaves per plant, leaf area index (LAI), and leaf-N concentration and content. The bulbing ratio, calculated as the ratio between the bulb's diameter and the false stem's diameter, was assessed in ten randomly selected plants per plot at 30, 60, 90, and 100 days after transplanting and at harvest. The date of BI was defined as the date when 50% of the plants had a bulbing ratio exceeding 2 (Brewster et al., 1987). Onion aboveground biomass and the number of leaves per plant were measured through destructive sampling of ten randomly selected plants per plot at transplanting, 30 and 60 days after transplanting, BI, and harvest. After dividing the plants into leaves, false-stems, and bulbs, all components were dried at 60°C for 48 h and weighed. LAI (m² of leaves m⁻² of soil) was assessed at BI as this index is a key determinant of crop yield (Dogliotti et al., 2021). LAI was estimated based on the leaf dry matter per plant, plant density, and the specific leaf area (SLA, cm² g⁻¹; Fang et al., 2019). SLA was estimated by sampling twelve circular fragments (16 mm diameter) of the middle part of fully developed onion leaves of ten randomly selected plants per plot. Leaf nitrogen concentration (%) was measured using the Kjeldahl method (Bremmer and Mulvaney, 1982) on a composite sample of ten randomly selected active and completely developed leaves per plot at 30 and 60 days after transplanting and at BI in both years, and at harvest in 2019. Leaf nitrogen content (kg leaf-N ha⁻¹) was estimated from leaf dry matter biomass, leaf N-concentration, and plant density.

In 2019, we observed differences in crop maturity at harvest. As a result, in 2020, we evaluated the proportion of green leaves and the percentage of collapsed foliage at harvest, indicative of bulb maturity (Brewster and Butler, 1989). The proportion of green leaves was assessed on ten randomly selected plants per plot using a four-level visual scale: 1: <40% green leaves, 2: 40–60%, 3: 60–80%, 4: >80%. The percentage of collapsed foliage was calculated as the ratio of the number of plants with collapsed foliage and the total number of plants in the eight central meters of the middle bed per plot, which was also used to assess yield.

2.3.4.3. Soil physical, chemical, and biological properties. Physical and chemical soil properties were measured in one composite sample per

plot consisting of twenty subsamples of top soil (0–20 cm) taken at the beginning of the CC (March) and the end of the onion growing season (December) in 2019 and 2020. After drying the soil samples and passing them through a 2 mm sieve, the following analyses were made: soil pH (1:2.5 soil:water and soil:KCl ratio), soil texture by hydrometer method (Forsythe 1975), SOC by Walkley-Black method (Nelson and Sommers, 1996), available P by Bray and Kurtz method (Bray and Kurtz, 1945), and exchangeable K by atomic emission spectrophotometry following ammonium acetate extraction (Isaac and Kerber, 1971). The bulk density of the top soil was estimated by taking undisturbed samples at 7–10 cm depth using three metal rings per plot (5 cm wide and 3 cm tall, Blake and Hartge, 1986) at CC sowing, onion transplanting, BI (only in 2019), and harvest. The SOC stock per ha was estimated considering 20 cm depth (2000 m³ soil ha⁻¹), the bulk density, and SOC (%). The relative active SOC (RASOC) was estimated according to Dogliotti et al. (2014). Soil temperature was measured with three Pendant MX2201 sensors per plot, located at 5, 15, and 25 cm depth, and measurements were taken every 15 minutes from transplanting to onion harvest. Temperature recordings were summarized as daily averages and were assigned to the early (1–15 September), mid (15–31 October), and late onion seasons (1–15 December) for both years.

Soil NO₃ and NH₄, and mineral N (calculated by adding NO₃ and NH₄) were measured in one composite sample per plot consisting of ten subsamples of top soil (0–20 cm) taken before onion transplanting, at 20, 50, and 70 days after transplanting, at BI, and before harvest. NO₃ and NH₄ were determined by colourimetric analysis (Doane and Horwath, 2003; Rhine et al., 1998, Appendix C). In 2019, soil basal respiration (Ohlinger et al., 1996, Appendix C) and potentially mineralisable nitrogen (PMN) (Kandeler, 1996, Appendix C) were assessed in each soil sample, while in 2020, these determinations were done only for the samples taken before transplanting and at BI. In 2019, the potential nitrification activity of ammonia oxidizers (PNA) (Rudisill et al., 2016), urease and dehydrogenase enzyme activity (Kandeler, 1996; Von Mersi and Schinner, 1991) were assessed as well (Appendix C).

2.3.5. Statistical analysis

We analyzed the effect of the tillage and NEM treatments on response variables using generalized linear mixed models. The response variables included: aboveground onion biomass, number of leaves, bulbing ratio, LAI at BI, total and marketable yield, bulb size, plant density and leaf-N concentration and content, CC aboveground biomass, CC C:N ratio, proportion of bare soil, and soil cover by the CC and residues, weed cover, weed aboveground biomass, soil mineral nitrogen, NH₄:NO₃ ratio, soil respiration, PMN, SOC, soil bulk density, and soil temperature. In 2019, we also assessed the effect of the treatments on PNA, urease, and dehydrogenase enzyme activity, and in 2020, the proportion of green leaves and the percentage of foliage collapse at harvest. The explanatory variables were tillage (RT or CT, whole plot factor), NEM application (presence or absence, split-plots factor), year (2019 and 2020), sampling date when the variables had repeated measures and their two-way interactions. The random effects were specified as follows: the tillage by block interaction as the whole plot error (1|Block:Tillage), and the NEM by tillage by block interaction as the pooled error (1|Block:Tillage:NEM) in which we included the date when repeated measures in time were modeled (1|Block:Tillage:NEM:DateNumber). We first evaluated models with "Year" (2019 or 2020) as a fixed effect. When interactions with "Year" were significant, separate analyses were conducted for 2019 and 2020. We used Gaussian or gamma error distributions for continuous variables and Poisson or negative binomial error distributions for count and proportion data (Appendix E). For response variables with many zeros, corrections were included in the model. Generalized linear mixed models were developed using the glmmTMB R-package (Brooks et al., 2017). Model residuals were checked using the DHARMA package in R (Hartig, 2022). For significant effects, least square means were adjusted, and the Tukey test for multiple comparisons was performed using the lsmeans R-package (Lenth,

2016). The R-packages "ggplot2" (Wickham, 2016), "ggpubr" (Kassambara, 2020), and "scales" (Wickham and Seidel, 2019) were used for data visualization.

2.4. On-farm experiments

In 2020, simplified experiments were conducted on two commercial farms approximately 25 km from the CRS. The farmers were participating in the support group of the project. Farm 1 comprised a conventionally managed system in which soil-improving practices had been applied for more than fifteen years (crop rotation with green manures, and four-year lucerne). Farm 2 had been a certified organic farm for five years. Onion was the main crop for more than ten years on both farms. The previous soil management was similar to the CRS, all with previous cultivation with CT. The soil type was similar to CRS (Mollic Vertisols, Hypereutric), with relatively high SOC and no soil-borne disease history. The farms had lower levels of SOC than CRS (Farm 1: 1.80%, Farm 2: 2.28%) and a similar topsoil bulk density (0.86 g cm⁻³). Detailed information is presented in Appendix B. On each farm, SOC, soil nutrient level, soil bulk density, CC biomass and quality, were similar across treatments at the start of the experiment ($p > 0.1$).

The treatments were discussed during a workshop and with each farmer in 2019. The experiment on Farm 1 consisted of two strips, one with CT and synthetic fertilizer (CT/SF) and NEM application. Three pseudoreplicate plots were placed within the strip to evaluate soil and crop performance. The second strip had RT with chicken manure (RT/CkM) management. Within the RT/CkM strip, NEM treatment (present+ or absent-) was inserted with a randomized design in three replicates. Tillage treatments were not replicated. Thus, there were three treatments (CT/SF/NEM+, RT/CkM/NEM+, RT/CkM/NEM-) with three pseudoreplicates per treatment on Farm 1 (Appendix F). The experiment on Farm 2 also consisted of two strips defined by the tillage system (RT and CT), and tillage treatments were not replicated. NEM treatment (present+ or absent-) was applied in each strip with a randomized block design with three replicates. Thus, there were four treatments (RT/NEM-, RT/NEM+, CT/NEM-, CT/NEM+) with three pseudoreplicates per treatment on Farm 2 (Appendix F). The variables assessed were similar to CRS except for soil temperature, which was only monitored in one RT and one CT plot per farm.

Crop sequences, CC species, and onion seedlings were similar to those at CRS. The on-farm experiments were managed according to farmers' practice, except for pre-defined nutrient management for each treatment and weed management, where herbicides were not allowed, and the research team did the weeding (Appendix F). Due to heavy rain, soil tillage in CT on Farm 2 was postponed, and transplanting took place 13 days later than on Farm 1 and the CRS. Moreover, Farm 2 had a water deficit after transplanting, and leaf-cutter ants caused severe plant damage in one of the three replicates.

3. Results

3.1. CRS on-station experiment

3.1.1. Cover crop biomass, quality, and soil cover

CC aboveground dry biomass in 2019 was 4.4 ± 1.1 Mg ha⁻¹ and had a C:N ratio of 31 ± 3 . At CC termination, foxtail millet was in a late development stage (starting seed maturation), and foxtail millet, cowpea, and weeds comprised 71%, 32%, and 7% of the biomass, respectively. In 2020, CC biomass was 7.1 ± 1.7 Mg ha⁻¹, with a C:N ratio of 21 ± 3 . Foxtail millet at milky ripe development stage constituted 94% of the biomass, and weeds constituted the remaining 6%. The drought in the summer of 2020 prevented the establishment of cowpea.

The soil coverage reached by the CC exceeded 80% in both years, and the bare soil was less than 15% (Fig. 2-A, B). In both years, RT had less than 20% of bare soil during the onion cycle. CT had 30% and 70% of bare soil until the end of September in 2019 and 2020, respectively. Soil

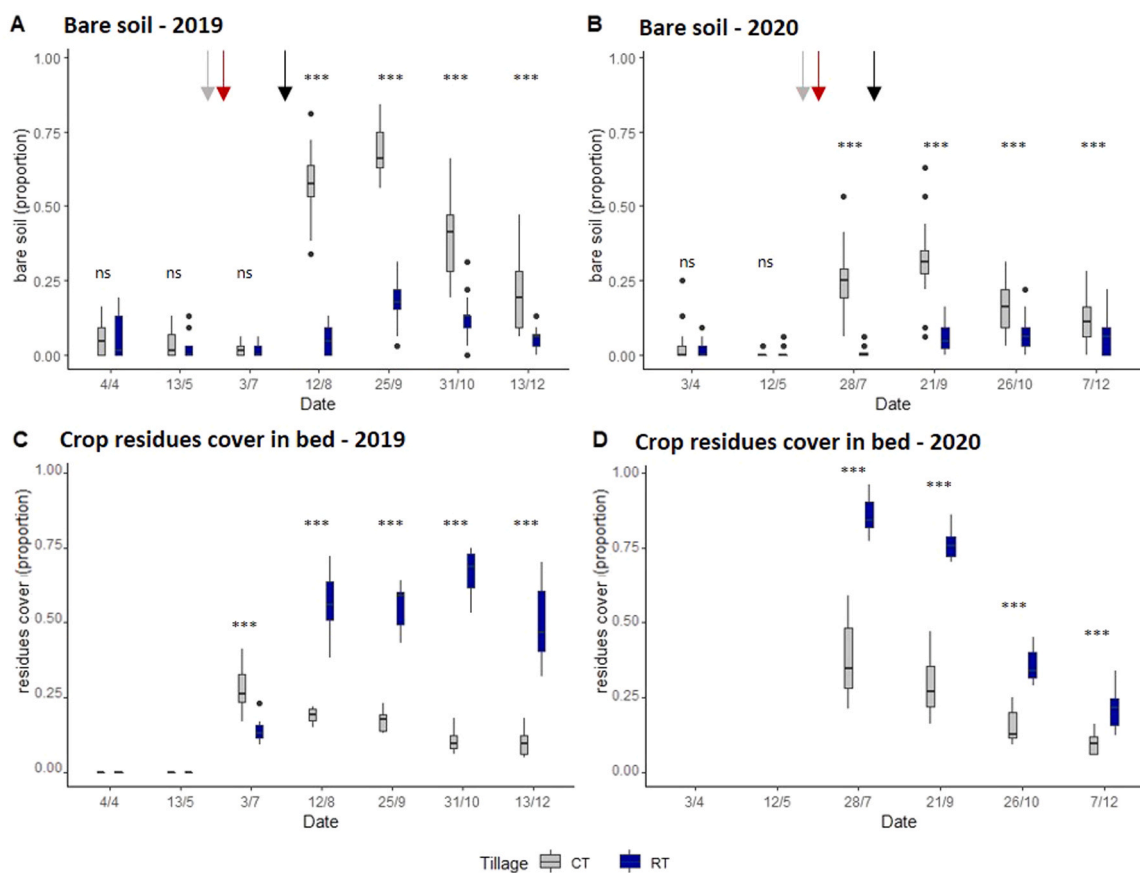


Fig. 2. Proportion of bare soil (top) and crop residues cover in the raised beds (bottom) during 2019 (left) and 2020 (right) for conventional tillage (CT, grey) and reduced tillage (RT, blue) during the cover crop and onion growing period. The grey arrow indicates the end of the cover crop cycle due to low temperatures, the red arrow indicates the start of tillage in the CT treatment, and the black arrow indicates onion transplanting. Asterisks indicate significant differences between tillage treatments per sampling date: **p* < 0.05; ** *p* < 0.01; *** *p* < 0.001, ns: non-significant. NEM application did not significantly influence vegetation cover.

residue cover during the onion cycle in RT was more than double that in CT (*p* < 0.001, Fig. 2-C, D). NEM application did not significantly affect soil cover (*p* > 0.05).

3.1.2. Onion yield and yield components

Total onion yield was affected differently by tillage each year (significant interaction *p* < 0.01). In 2019, there was no significant difference between tillage treatments for total yield, while in 2020, total yield in RT was 33% lower than in CT (*p* < 0.001). Total yield was not significantly affected by NEM. Marketable yield showed a significant interaction between year and tillage (*p* < 0.05) and between tillage and NEM (*p* < 0.05). In 2019, the marketable yields of CT and RT were comparable without NEM (around 7.5 Mg ha⁻¹), while with NEM, CT had around

33% higher marketable yield than RT. In 2020, marketable yield in CT was 36% higher than in RT (*p* < 0.001), and NEM did not have a significant effect. The differences in total and marketable yields were explained by bulb size and not by the number of bulbs per ha (Table 1).

3.1.3. Onion growth and development

LAI at BI was greater in 2020 than in 2019 (*p* < 0.001), and there was a significant interaction between year and tillage (*p* < 0.01, Table 1). In 2019, no significant differences among treatments were found. In 2020, LAI at BI in RT was 27% lower than CT (*p* < 0.05). The LAI differences were related to plant size and not to plant density (Table 1, Fig. 3), and the plant size differences were related to plant aboveground biomass and not to the number of leaves per plant (Fig. 3). There was no

Table 1

Total and marketable onion yield, bulb size, plant density, and leaf area index at bulb initiation (LAI at BI) per treatment for the two experimental years at the CRS station.

Year	Tillage	NEM application ¹	Total yield (Mg ha ⁻¹)	Marketable yield (Mg ha ⁻¹)	Average bulb fresh weight (g)	Plant density at harvest (plants m ⁻²)	LAI at BI (m ² leaf m ⁻²)
2019	Conventional	No	10.5 ± 5.0 a	8.8 ± 6.2 b	53 ± 26 b	20 ± 2 a	0.56 ± 0.25 a
		Yes	12.2 ± 5.0 a	11.2 ± 5.6 a	61 ± 25 a	20 ± 2 a	0.53 ± 0.15 a
	Reduced	No	10.0 ± 3.0 a	8.3 ± 3.7 b	47 ± 10 b	21 ± 4 a	0.55 ± 0.13 a
		Yes	9.5 ± 2.5 a	7.5 ± 3.1 b	41 ± 10 b	23 ± 2 a	0.54 ± 0.15 a
2020	Conventional	No	15.3 ± 1.5 a	14.7 ± 1.5 a	89 ± 4 a	17 ± 2 a	1.02 ± 0.22 a
		Yes	15.2 ± 2.5 a	14.7 ± 2.7 a	87 ± 20 a	18 ± 2 a	1.13 ± 0.34 a
	Reduced	No	11.4 ± 1.1 b	10.5 ± 1.2 b	61 ± 9 b	19 ± 3 a	0.90 ± 0.18 b
		Yes	9.3 ± 1.8 b	8.4 ± 2.0 b	55 ± 7 b	17 ± 2 a	0.75 ± 0.15 b

¹ Native effective microorganisms

Different letters indicate significant differences among treatments within a year (*p* < 0.05).

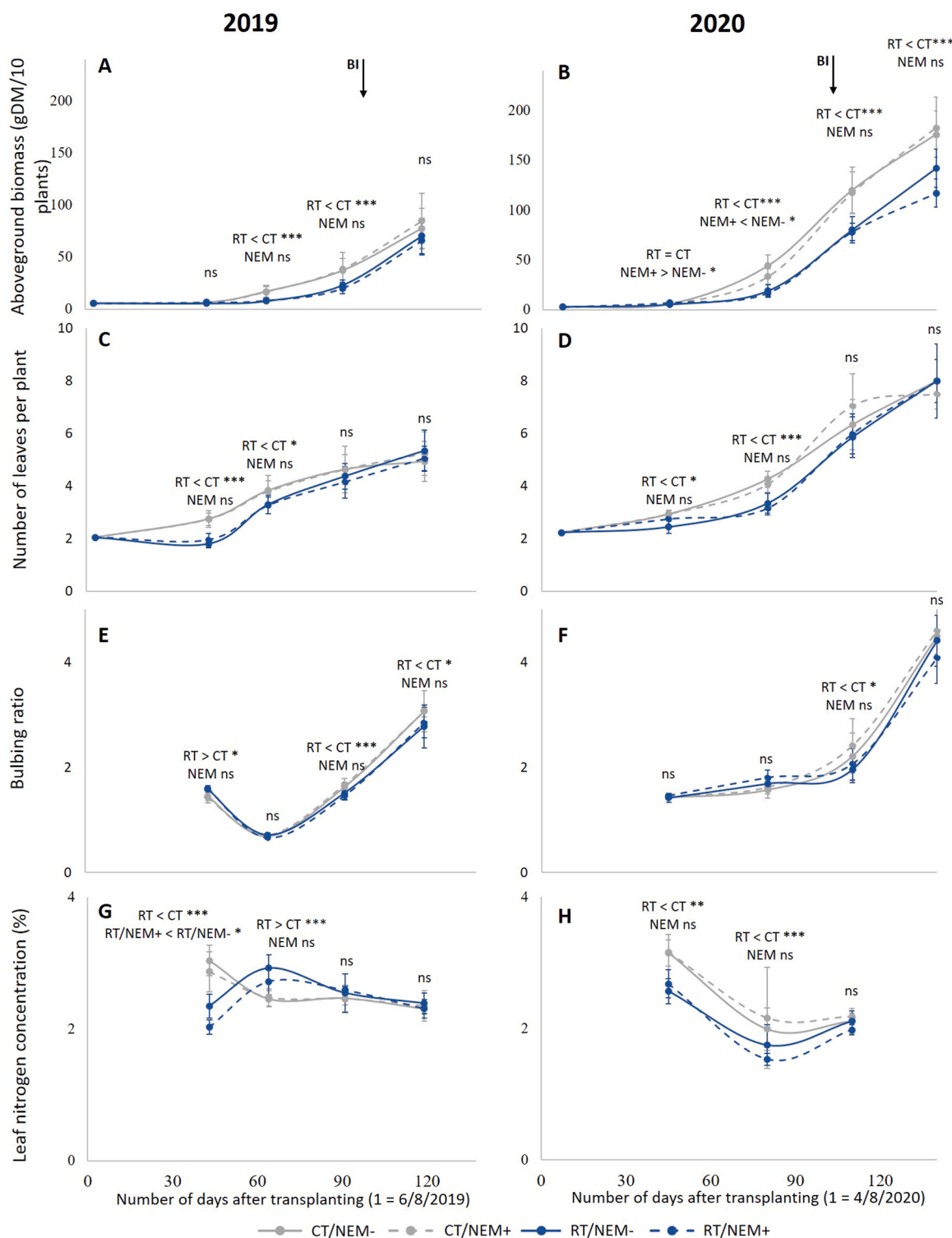


Fig. 3. Aboveground biomass, number of leaves per plant, bulbing ratio, and leaf nitrogen concentration of onion during the 2019 (left) and 2020 (right) growing seasons for conventional tillage (CT, grey) and reduced tillage (RT, blue) without NEM (solid) and with NEM (dashed). Black arrows indicate bulb initiation. Asterisks indicate significant differences between treatments per sampling date: * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; ns: non-significant.

significant effect of NEM on LAI.

Aboveground onion biomass and growth rate after day 45 was higher in 2020 than in 2019, and there was a significant interaction between year and tillage (Fig. 3-A and B). In 2019, CT had higher aboveground biomass than RT at two months after transplanting and at BI ($p < 0.001$), and in 2020, from two months after transplanting to harvest ($p < 0.001$). In 2019, there was no significant effect of NEM on aboveground onion

biomass. In 2020, there was an interaction between NEM and date ($p < 0.01$). One month after transplanting, onions with NEM had a higher biomass than those without NEM ($p < 0.05$). In contrast, two months after transplanting, onions without NEM had a higher biomass than onions with NEM ($p < 0.05$, Fig. 3-A and B).

Although onions in CT and RT had a similar crop development in terms of number of leaves per plant at BI and at harvest, CT showed a

greater number of leaves in the initial two-month period after transplanting than RT ($p < 0.05$ and < 0.001 depending on the date and year, Fig. 3-C and D). The bulbing ratio in RT was lower than in CT at BI ($p < 0.001$ in 2019 and $p < 0.05$ in 2020, Fig. 3-E and F), indicating a delay in crop development. Moreover, in 2020, RT had a higher proportion of green leaves ($p < 0.05$) and a lower percentage of collapsed foliage at harvest than CT ($p < 0.05$, Appendix G), indicating that the onion crop in RT was in an earlier development stage at harvest than in CT.

Leaf-N concentration and content in the onion leaves were influenced by tillage treatment and date. In 2019, one month after transplanting, the leaf-N concentration in RT onion was lower than in CT onion ($p < 0.001$), but two months after transplanting, this difference was reversed ($p < 0.001$, Fig. 3-G and H). Leaf-N content of RT was lower than that of CT from one month after transplanting until BI (Appendix G). In 2020, RT onion had a 20–25% lower leaf N-concentration than CT until BI ($p < 0.01$, Fig. 3-H), and lower leaf-N content from two months after transplanting (Appendix G). NEM only affected onion leaf N-concentration one month after transplanting in 2019, where RT/NEM+ had a lower leaf-N concentration than RT/NEM- ($p < 0.05$, Fig. 3-G).

3.1.4. Weed pressure

During the CC cycle, weed soil cover was always lower than 15% and was often virtually absent. However, after CC senescence and before weeding in RT or tillage in CT (July), weed soil cover increased to more than 25% (Fig. 4-A and B).

Weed biomass and soil covered by weeds at onion transplanting in

both years were higher in RT than in CT (Fig. 4). From transplanting to two or three months after transplanting, RT still had higher weed pressure than CT despite manual weeding applied to both tillage systems (Fig. 4). After that, weed variables did not differ between RT and CT, except at BI in 2019 when weed biomass was higher in CT than in RT ($p < 0.05$, Fig. 4).

In 2019, three weeding events for CT and RT were carried out during the onion cycle, requiring 740 h human labour ha^{-1} . In 2020, RT required four weeding interventions and 1390 h ha^{-1} , while CT required three weeding interventions and 620 h ha^{-1} .

In 2019, the main weed species were *Stachys arvensis* (Lamiaceae), *Bowlesia incana* (Apiaceae), and *Echinochloa crus-galli* (Poaceae). In 2020, the main weed species were *Stachys arvensis* (Lamiaceae), *Stellaria media* (Caryophyllaceae), *Digitaria sanguinalis* (Poaceae) and *Bowlesia incana* (Apiaceae) (Appendix H).

3.1.5. Soil physical, chemical, and biological properties

The RT treatment had lower bulk density than CT (0.898 vs 0.961 g cm^{-3} , $p < 0.01$, Appendix I). Soil temperature showed season-specific responses to the tillage treatment each year ($p < 0.01$). In 2019, soil temperatures were 2°C and 1°C lower than in 2020 in early and mid-season, respectively ($p < 0.001$, Appendix I). In 2019, RT had 0.5°C lower mean, minimum, and maximum soil temperatures at 5 cm depth than CT at late season ($p < 0.05$), while in 2020, RT had 0.5°C lower temperatures than CT at 5 cm ($p < 0.05$) and 10 cm depth in early season ($p < 0.01$, Appendix I).

During the two-year experiment, the SOC was not significantly

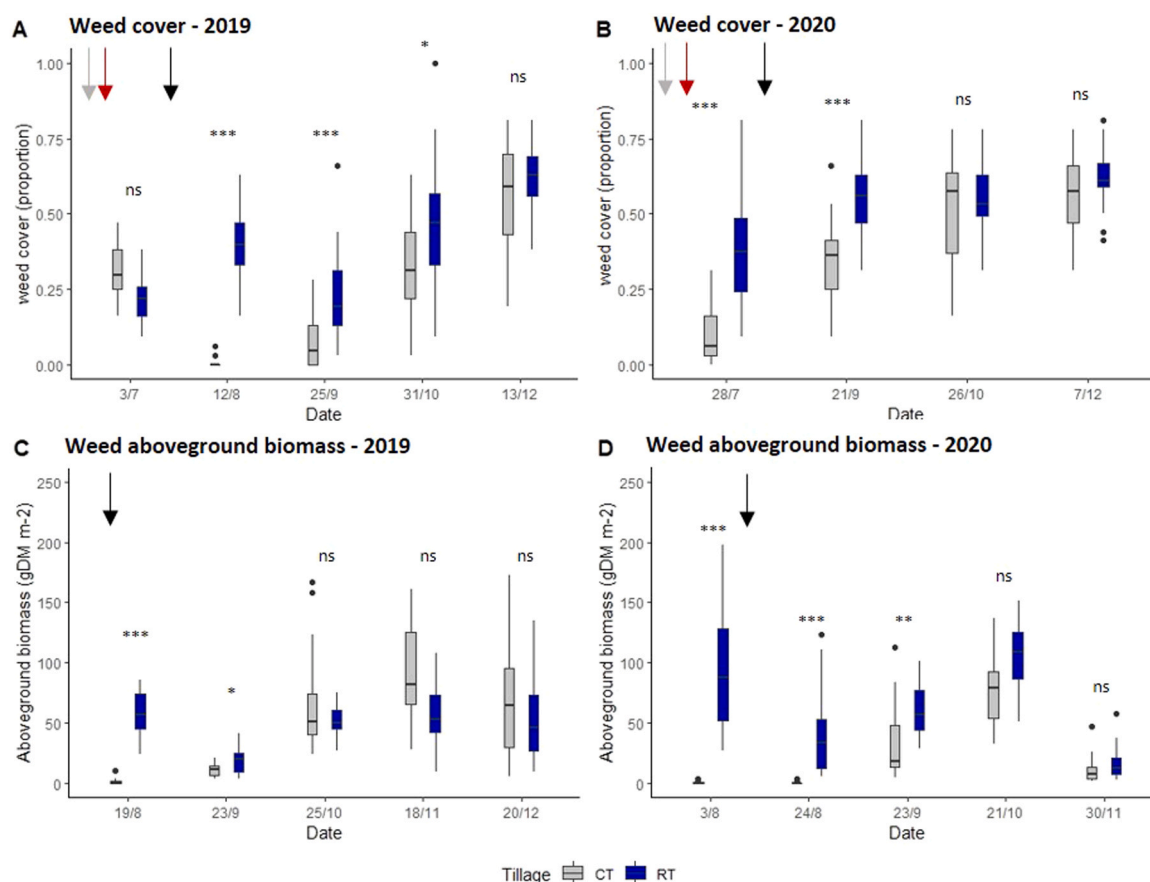


Fig. 4. Proportion of soil covered by weeds (top) and weed aboveground biomass (bottom) during the onion cycle in 2019 (left) and 2020 (right) for conventional tillage (CT, grey) and reduced tillage (RT, blue). The grey arrow indicates the end of the cycle of the cover crop, the red arrow indicates the starting of tillage in the CT treatment, and the black arrow indicates onion transplanting. Sampling dates of weed biomass (19/8/2019 and 3/8/2020) and proportion of weed cover assessments (12/8/2019 and 28/7/2020) were performed after soil tillage in the CT treatment and before the first manual weeding in the RT. Asterisks indicate significant differences between tillage treatments per sampling date: * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$, ns: non-significant. There was not significant effect of NEM application treatment.

influenced by tillage, NEM application, or sampling date (Appendix I). RT tended to have higher respiration rates than CT in both years and all sampling dates ($p=0.056$, Appendix I) and had higher PMN at BI in 2019 ($p<0.05$, Appendix I). In 2019, PNA at transplanting and BI were higher in RT than in CT ($p<0.01$), RT also had higher urease enzyme activity ($p<0.05$) and a tendency to have higher dehydrogenase enzyme activity at transplanting than CT ($p<0.1$, Appendix I). No significant effect of NEM on the assessed biological parameters was identified.

The tillage and NEM treatments did not significantly influence the total mineral nitrogen in both years. Still, the level of soil mineral nitrogen at each sampling date in 2019 was around five times lower than in 2020 ($p<0.001$, Fig. 5-A and B). In 2019, total mineral nitrogen peaked after transplanting (19 mg kg^{-1}) and remained around 11 mg kg^{-1} until harvest (Fig. 5-A). In 2020, total mineral nitrogen peaked after transplanting (100 mg kg^{-1}) and oscillated around 62 mg kg^{-1} until harvest (Fig. 5-B). The $\text{NH}_4\text{:NO}_3$ ratio was always below 2, but in 2019 it was more than double that in 2020 ($p<0.001$, Appendix I), and in 2020, the ratio in RT was higher than in CT ($p<0.001$, Appendix I).

3.2. Comparison between CRS and on-farm experiments

The on-farm experiments confirmed some of the findings from the CRS experiment (Table 2): i. leaf-N content in early stages of the onion crop and LAI at BI were lower for RT than for CT; ii. RT resulted in a delay in bulbing ratio and crop maturity; iii. soil mineral nitrogen did not differ among treatments, but RT had a higher $\text{NH}_4\text{:NO}_3$ ratio than CT; iv. soil biological activity tended to be higher in RT than in CT; v. weed biomass and weed soil cover in the early stages of the onion crop and weeding workload were higher in RT than in CT; vi. soil cover by CC residues was higher in RT than CT during the entire cycle.

However, there were also contrasting findings in the on-farm and CRS experiments (Table 2): i. RT had a higher bulk density than CT on both farms, while this was the other way around in the CRS; ii. on Farm 1, soil mineral nitrogen in the CT/SF treatment was more than double the levels of the RT/CkM and more than any treatment on Farm 2 and CRS; iii. on Farm 1, yield of CT/SF was comparable to RT/CkM/NEM- and LAI levels were higher than in the other locations; iv. Farm 1 had a lower weed pressure compared to CRS and Farm 2. The results of commercial farms are presented in Appendix J.

4. Discussion

In this study, we assessed the effect of CC-RT vs CC-CT in

combination with NEM application on the performance of onion crops, N status, and weed pressure without applying herbicides or synthetic fertilizers at three locations in the south of Uruguay. We report four key findings. First, onion growth and yields were generally low. Yields were lower in 2019 than in 2020 and lower in RT than in CT. Low yields were associated with relatively low leaf-N concentrations in the early stages of crop development. Second, soil mineral N was lower in 2019 than in 2020 and did not significantly differ between treatments, while the $\text{NH}_4\text{:NO}_3$ ratio was higher in 2019 than in 2020 and higher in RT than in CT. Soil physical properties were not limiting crop growth in any treatment, and biological activity was higher in RT than in CT. Third, although RT resulted in high soil residue cover, weed pressure in the early crop stages was higher than in CT, which was reversed at later stages. Both RT and CT required manual weeding during the onion cycle, but the workload was higher in RT than in CT. Fourth, the NEM application did not significantly affect most crop, weed, and soil variables.

4.1. Effect of tillage on onion growth, development and yield

All treatments in both years had relatively low yields compared to the average (26 Mg ha^{-1}) and attainable yields (45 Mg ha^{-1}) for the region (Dogliotti et al., 2021). These low yields were related to low crop growth rates before BI, evidenced by LAI levels at BI from 0.53 to $1.13 \text{ m}^2 \text{ leaf m}^{-2}$, while at least a LAI of 1 and $2 \text{ m}^2 \text{ leaf m}^{-2}$ is needed to achieve the average and attainable yield, respectively (Dogliotti et al., 2021). CT treatments in 2020 had a LAI at BI above $1 \text{ m}^2 \text{ leaf m}^{-2}$, but the total yield was still lower than the regional average, suggesting that the crop growth rate after BI was also low.

The low crop growth rate before BI can be explained by the observed leaf-N concentrations below the critical threshold of 4% at an early crop stage (Brewster et al., 1987; Geisseler et al., 2022; Maynard and Hochmuth, 2007). Nitrogen deficiency reduces gross assimilation, growth rates, and leaf area expansion (Geisseler et al., 2022; Brewster and Butler, 1989). Moreover, in line with our results, N shortage during early stages has a negative effect on crop development, lowering leaf initiation rate and bulb formation (Brewster and Butler, 1989). Thus, N deficiencies may explain the low yield levels of the CRS experiment in both years, the better onion performance in 2020 than in 2019, and the lower yield in RT than CT in CRS in 2020. The better crop performance in the CRS experiment in 2020 than in 2019 could also be partially explained by the earlier appearance and higher incidence and severity of downy mildew in 2019 than in 2020 (Appendix G).

A promising result was that in CRS and on Farm 1 in 2020, RT reached LAI levels close to 1, which could have allowed yield levels of

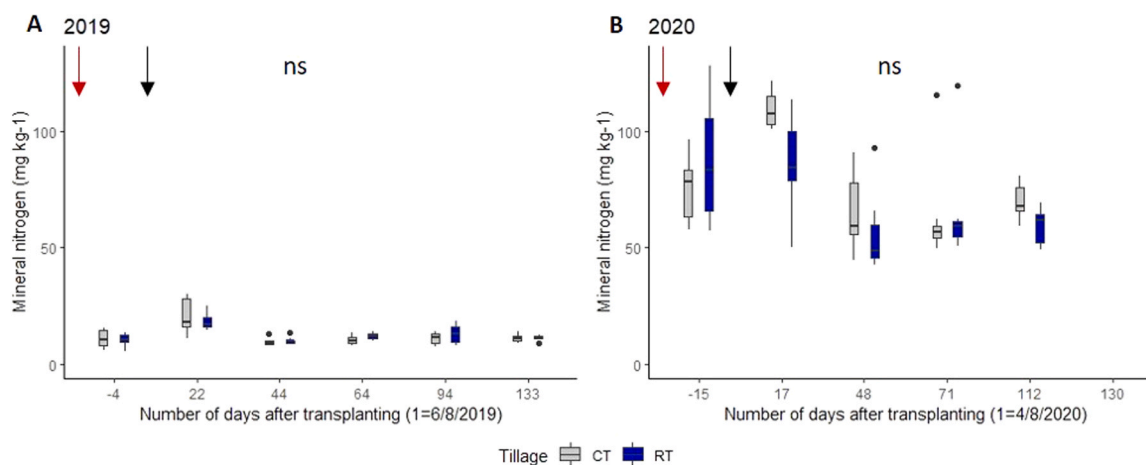


Fig. 5. Soil mineral nitrogen during the onion cycle in 2019 (left) and 2020 (right) for conventional tillage (CT, grey) and reduced tillage (RT, blue). The red arrow indicates tillage in the CT treatment, and the black arrow indicates onion transplanting. Asterisks indicate a significant difference between tillage treatments each year: * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$, ns: non-significant. There was not significant effect of NEM application treatment.

Table 2

Summary of results on the cover crop (CC), soil cover, onion yield, onion crop growth and development, weed pressure and soil properties at the CRS Experimental Station in 2019 and 2020, and the two commercial farms in 2020. For each variable, significant differences among treatments are indicated (RT: reduced tillage, CT: conventional tillage; NEM+: with NEM, NEM-: without NEM), and NS indicates non-significant relationships. Mean values and (standard deviations) are shown.

	Location and year	CRS 2019	CRS 2020	Farm 1 2020	Farm 2 2020
	Treatments	RT/NEM+, RT/NEM-, CT/NEM+, CT/NEM-	RT/NEM+, RT/NEM-, CT/NEM+, CT/NEM-	RT/CkM/NEM+, RT/CkM/NEM-, CT/SF/NEM+ ^a	RT/NEM+, RT/NEM-, CT/NEM+, CT/NEM-
SOC and cover crop	Soil organic carbon (%)	2.55 NS	2.55 NS	1.80 NS	2.26 NS
	RASOC ^b	0.62 NS	0.62 NS	0.35 NS	0.52 NS
	CC Biomass (Mg ha ⁻¹)	4.4 (1.1) NS	7.1 (1.7) NS	3.9 (0.3) NS	4.3 (0.5) NS
	CC C:N ratio	31 ± 3 NS	21 (3) NS	15 (1) NS	26 (1) NS
	Developmental stage at termination	Mature grains	Milky ripe grains	Anthesis	Milky ripe & mature grains
Soil cover	Bare soil at BI ^c (%)	RT < CT 12 (7) vs. 39 (14)	RT < CT 7 (6) vs. 16 (8)	RT/CkM < CT/SF 9 (6) vs. 39 (12)	RT < CT 24 (7) vs. 58 (11)
	Residues cover in bed at BI ^c (%)	RT > CT 67 (8) vs. 10 (4)	RT > CT 36 (6) vs. 15 (6)	RT/CkM > CT/SF 69 (13) vs. 7 (1)	RT > CT 43 (9) vs. 10 (3)
	Total yield (Mg ha ⁻¹)	NS 10.6 (3.7)	RT < CT 10.3 (1.8) vs. 15.3 (1.9)	RT/CkM/NEM+ < other two 14.0 (1.5) vs. 15.9 (1.6)	RT < CT 2.9 (1.0) vs. 6.2 (1.0)
Yield and crop growth and development	Marketable yield (Mg ha ⁻¹)	RT & CT/NEM- < CT/NEM+ 8.2 (4.1) vs. 11.2 (5.6)	RT < CT 9.4 (3.2) vs. 14.7 (2.0)	RT/CkM/NEM+ < other two 13.0 (1.7) vs. 15.2 (1.7)	RT < CT 1.7 (1.1) vs. 5.2 (1.1)
	LAI at BI ^c m ² leaf m ⁻²	NS 0.54 (0.16)	RT < CT 0.83 (0.18) vs. 1.1 (0.3)	RT/CkM < CT/SF 1.1 (0.1) vs. 1.4 (0.1)	RT < CT 0.3 (0.1) vs. 0.4 (0.1)
Weed pressure	Leaf-N after transplanting (%)	RT/NEM+ < RT/NEM- < CT 2.0 (0.1) vs. 2.3 (0.2) vs. 3.0 (0.3)	RT < CT 2.6 (0.2) vs. 3.2 (0.2)	NS 2.8 (0.2)	NS 2.7 (0.3)
	Leaf-N at BI ^c (%)	NS 2.6 (0.2)	RT < CT 2.1 (0.1) vs. 2.1 (0.1)	RT/CkM < CT/SF 2.3 (0.2) vs. 2.7 (0.1)	NS 2.1 (0.1)
	Bulbing ratio at BI ^c	RT < CT 1.5 (0.2) vs. 1.8 (0.2)	RT < CT 2.0 (0.3) vs. 2.3 (0.5)	RT/CkM < CT/SF 2.5 (0.5) vs. 2.9 (0.5)	NS 2.4 (0.5)
	Foliage collapse at harvest (%)	Not assessed	RT < CT 4 (2) vs. 8 (4)	RT/CkM < CT/SF 8 (1) vs. 14 (2)	Not assessed
	Green leaf at harvest (%)	Not assessed	RT > CT 83 (7) vs. 70 (15)	RT/CkM > CT/SF 77 (5) vs. 50 (26)	Not assessed
	Weed biomass before treatments (g dm m ⁻²)	NS 57 (19)	NS 95 (49)	NS 46 (36)	NS 167 (46)
	Weed biomass after transplanting (g dm m ⁻²)	RT > CT 19 (10) vs. 11 (6)	RT > CT 38 (32) vs. 1 (1)	RT > CT 7 (7) vs. 1 (3)	RT > CT 19 (11) vs. 6 (8)
	Weed biomass at BI ^c (g dm m ⁻²)	RT < CT 51 (13) vs. 62 (39)	NS 93 (30)	NS 11 (10)	RT > CT 34 (14) vs. 8 (10)
	Number and time (h ha ⁻¹) of weeding interventions	NS Three 725 vs 750	RT > CT Four vs three 1390 vs 620	RT > CT Three vs two 480 vs 255	RT > CT Three 1170 vs 560
	Soil chemical, physical and biological properties	Mineral N after transplanting mg kg ⁻¹	NS 19 (6)	NS 100 (20)	RT/CkM < CT/SF 85 (26) vs. 228 (40)
NH ₄ :NO ₃ ratio after transplanting		NS 1.7 (0.6)	RT > CT 0.4 (0.1) vs. 0.2 (0.1)	NS 0.4 (0.1)	NS 0.5 (0.3)
Mineral N at BI ^c mg kg ⁻¹		NS 12 (3)	NS 64 (21)	RT/CkM < CT/SF 53 (10) vs. 305 (78)	NS 49 (9)

(continued on next page)

Table 2 (continued)

Location and year	CRS 2019		CRS 2020		Farm 1 2020		Farm 2 2020	
NH ₄ :NO ₃ ratio at BF ^a	NS	RT > CT	RT > CT	RT > CT	RT/CKM > CT/SF	NS	NS	
Respiration at BF ^c mg C-CO ₂ kg ⁻¹	1.2 (0.6)	RT > CT	1.2 (0.6) vs. 0.9 (0.5)	RT > CT	0.3 (0.1) vs. 0.1 (0.0)	0.1 (0.0)	0.1 (0.0)	
Potentially mineralizable N at BF ^c mg N. g ⁻¹ d ⁻¹	5.3 (1.3) vs. 4.6 (1)	RT > CT	5.3 (1) vs. 4.6 (1)	NS	RT/CKM > CT/SF	NS	NS	
Bulk density before harvest g cm ³	8.3 (1.7) vs. 6.3 (1.5)	RT < CT	6.1 (1.2)	RT < CT	35.1 (10.2) vs. 21.7 (3.4)	32.0 (7.0)	32.0 (7.0)	
Mean temperature 5 cm depth ^d °C	0.98 (0.15) vs. 1.06 (0.14)	RT < CT at late season	0.89 (0.08) vs. 0.94 (0.11)	RT < CT at early season	RT/CKM > CT/SF	5.7 (1.1)	5.7 (1.1)	
	23.6 vs. 24.1		14.5 vs. 15.0		RT/CKM > CT/SF	RT > CT	RT > CT	
					0.97 (0.07) vs. 0.88 (0.02)	1.00 (0.11) vs. 0.94 (0.08)	1.00 (0.11) vs. 0.94 (0.08)	
					RT < CT at late season	RT < CT at late season	RT < CT at late season	
					22.5 vs. 23.0	24 vs. 25.5	24 vs. 25.5	

^a In Farm 1: CkM: chicken manure, SF: synthetic fertilizer.

^b Relative Active Soil Organic Carbon (RASOC) = ((Actual SOC – Min SOC)/(Max SOC – Min SOC)) * 100 (Dogliotti et al., 2014)

^c BF: bulb initiation.

^d Early season: 1–15 September, Late season: 1–15 December.

around 25 Mg ha⁻¹ (Dogliotti et al., 2021). However, these yields in RT were not achieved, most likely because the crop was harvested prematurely since its development was delayed compared to the CT treatment. We estimate that the RT onions could have continued to grow for at least one or two more weeks. Therefore, the onion yields in RT are conservative, and higher yields can be expected in more optimized RT systems.

4.2. Effect of tillage on soil physical, chemical, and biological properties

Unfavorable soil physical conditions may limit the effectiveness of RT (Carr et al., 2013; Peigné et al., 2007). The lower bulk density in RT than in CT at the CRS experiment suggests that relatively high SOC and RASOC levels allow soil structure and porosity to be maintained despite not being tilled. In contrast, intensive soil tillage, such as in CT, increases macro porosity in the short term but deteriorates soil structure and increases SOC mineralization, which in the mid-to-long term result in more compaction (Weil and Brady, 2016). The two commercial farms showed a contrasting pattern compared to the CRS by having higher soil bulk densities in the RT treatment than for CT. This finding may be related to the lower SOC and RASOC levels of the soils at the farms than at the CRS, which may point to a lower soil physical quality or resilience (Hoffland et al., 2020), and highlights that good soil quality is needed for the effective implementation of RT.

Nevertheless, on both farms, the soil bulk density was below 1.1 g cm⁻³ and should not limit crop growth on this type of soil (USDA, 1999). In addition, farmers identified the "softness" of the soil as a positive and unexpected result of the experiment: "Contrary to what I expected to happen, the soil was not tight in the RT. It was softened and better than in the CT (...); beyond that, it is even a good result for the soil quality. It is also good because, for planting by hand, the soil has to be soft; otherwise, nobody will do it".

Soil biological activity was greater in RT than in CT, which aligns with previous studies (Arbolea et al., 2012; Carr et al., 2013). The urease activity is a good index of soil quality as it is closely related to soil organic matter, N cycling, and the regulation of N supply to plants (Adetunji et al., 2017). The higher urease activity found in 2019 in RT compared to CT may point to a higher potential organic matter mineralization rate in RT than CT. However, biological activity was measured under laboratory-controlled conditions, and *in situ* mineralization on the field depends on actual soil moisture and temperature, which was affected by tillage and could explain the observed crop N limitations.

RT had significantly lower topsoil temperatures than CT in the late season of 2019 and early season of 2020, which aligns with the findings of Coolman and Hoyt (2018) and Jokela and Nair (2016). These relatively low temperatures in RT can explain the delay in onion growth and development by a direct effect on the crop development rate (Lancaster et al., 1996) and by an indirect effect on the N availability for the crop via a reduction in root activity and N release by soil biota (Ciaccia et al., 2015).

The higher soil mineral N in 2020 than in 2019 at the CRS may be explained by a combination of effects. First, the relatively low C:N ratio of the CC, the relatively early soil tillage, and the compost tea application in 2020 may have reduced N immobilization by microorganisms and accelerated N mineralization (Hodge et al., 2000; Masunga et al., 2016; Mooshammer et al., 2014). Second, the higher precipitation in 2019 than in 2020 may have increased N-leaching (Ciaccia et al., 2015). Third, the lower soil temperatures at early and mid-onion seasons in 2019 compared to 2020 may have reduced soil organic matter mineralization (Luce et al., 2011). Fourth, the positive cumulative effect of the two years of chicken manure and CC may have contributed to a higher N level in 2020 than in 2019. Although the higher level of soil mineral N in 2020 increased N uptake by the onion crop, leading to higher LAI and aboveground biomass than in 2019, leaf-N concentrations kept below the critical thresholds limiting crop growth and development (Brewster et al., 1987; Geisseler et al., 2022; Maynard and Hochmuth, 2007).

We did not find significant effects of tillage on soil mineral N levels.

However, RT negatively affected the onion leaf-N concentration before BI (Fig. 3). Possibly, the timing of the soil mineral N measurements done every 20 or 30 days was not effective in tracking the extremely dynamic soil mineral N pool (Hodge et al., 2000). Therefore, although our data do not allow us to confirm it, the N immobilization effect after the CC might have lasted longer in the RT than in CT, probably due to a more limiting soil mineral N availability (Terrazas et al., 2016). These results highlight the complexity involved in the strong dynamics of organic matter mineralization and nutrient availability (Geisseler et al., 2022; Hodge et al., 2000; Masunga et al., 2016), underlining the challenge of matching the supply and demand of N under CC-RT without using synthetic fertilizers.

We found differences in the $\text{NH}_4\text{:NO}_3$ ratio between years and tillage treatments that may be related to the weather conditions (Appendix D), soil temperature and moisture conditions (Alliaume et al., 2017; Puerta et al., 2019; Wacker et al., 2022). However, the values were always within the range of 1:3 and 3:1 reported for good onion crop performance (Abbès et al., 1995; Gamiely et al., 1991).

On Farm 1, synthetic fertilizer (urea) was applied in CT/SF like for most onion farms in the region (Scarlato et al., 2022), resulting in three to six times higher soil mineral N in CT/SF than in RT, mainly in the form of NO_3 . However, onion yields of the RT/CkM/NEM- and CT/SF treatments were comparable (Table 2 and Appendix J). High mineral N, particularly high NO_3 conditions, indicate inefficiencies and pose an environmental risk because of N-leaching (Terrazas et al., 2016) and N_2O emission (Butterbach-Bahl et al., 2013). The results of Farm 1 evidence the environmental risk of the current most common management in the region based on CT and synthetic fertilizers. At the same time, it highlights the positive contribution that CC-RT, in combination with organic amendments, may have to reduce this environmental risk.

4.3. Effect of cover crop and tillage on weed pressure and soil cover

The CC of foxtail millet and cowpea resulted in an aboveground biomass of 4–7 Mg DM ha^{-1} and more than 80% of soil cover following a growing period of fewer than four months during the end of summer and autumn, which can be considered good performance for the study region, and can reduce soil erosion and increase soil organic matter (Alliaume et al., 2013; García De Souza et al., 2011; Gilsanz, 2012). The CC was naturally terminated by low temperatures in June, around one and a half months before onion transplanting. This provided sufficient time to prepare the onion transplanting while eliminating the need for herbicide applications to terminate the CC. Farmers appreciated this outcome: *"I am not completely sure if I will continue trying the reduced tillage technology yet, but I am sure I will continue using this cover crop in the system. It has a great performance, I do not need to use herbicide, and it is easy to manage before installing the onion (...). In general, it is not easy to include a cover crop in summer and before the onion because you need the land in summer for other crops and in general it is difficult to prepare the soil in autumn. But this option has shown that it is a very good and viable one."*

Despite the good CC performance, the CC residue in RT was ineffective in suppressing weeds. Consequently, RT required more weeding labour than CT and has likely increased competition for N with the onion crop in the early stages of crop development. While previous studies found that a mulch biomass of around 5 Mg ha^{-1} can effectively suppress weeds (Altieri et al., 2011; Leavitt et al., 2011), these studies focused on winter CC followed by summer crops (tomato, beans, sweet pepper, zucchini), which are competitive with weeds. Other studies showed that at least 8 Mg ha^{-1} of mulch biomass and a mulch thickness of 10 cm were needed to effectively suppress weeds (Teasdale and Mohler, 2000).

The differences in weed pressure between sites underline the relevance of the seed bank on the feasibility of CC-RT technology. In situations of high weed pressure, such as CRS and Farm 2, holistic and long-term strategies have to be considered beforehand to reduce soil weed seed bank, such as crop rotation with winter and summer green manure

and pastures, mechanical weeding, and false sow beds (Chikowo et al., 2009; Portela, 2008). The CC-RT technology could be complemented by adding external residues to increase the mulch thickness and the duration of soil cover (Teasdale and Mohler, 2000). The CC species selection and/or management could also be improved to achieve higher biomass, for example, by using highly productive C4 species and an extended growing season. Still, care is needed to trade-offs between N-immobilization and CC biomass. Finally, other mechanisms of weed suppression, such as the use of CC with allelopathic and inhibitory compounds, could be included (Shirgapure and Ghosh, 2020).

4.4. Effect of NEM on onion crop system

NEM application had no significant effect on any variable related to soil properties and a few related to crop performance. We expected that NEM would enhance the mineralization of organic materials and the associated nutrient availability for the crop (Higa and Wididana, 1991). In contrast, NEM application in the RT system in 2019 at the CRS resulted in a significantly lower leaf-N concentration 40 days after transplanting. On Farm 1, it led to significantly lower total and marketable onion yields than RT without NEM application. These unexpected findings may be explained by competition for N between plants and microorganisms in an N-limiting environment (Hodge et al., 2000). However, further research is needed to elucidate the mechanisms underlying these results.

4.5. Lessons learned and future steps

The CC-RT system tested in this study could limit bare soil cover to below 20% during the onion cycle, did not impose any constraints on soil bulk density for crop growth, and achieved good CC performance without using synthetic fertilizers and herbicides. However, the low onion yields, their link to nitrogen deficiencies and high weed pressure in early stages, and the site-specific results indicate that further improvements are needed.

The main benefits of CC-RT are expected and should be assessed in the long term, especially regarding soil health. Those benefits will result from organic matter accumulation and greater microbial biomass and activity, positively influencing soil biological, chemical, and physical attributes (Carr et al., 2013; Lv et al., 2023). Consequently, to continue testing and improving the application of the RT technology, it is necessary to set up a long-term research platform.

Long-term research approaches that seek to generate appropriate technological alternatives require working in application contexts and involving end-users (Doré et al., 1997; Doré et al., 2011; Milleville, 1993), in our case, farmers and technical advisers. In this context, participatory approaches promote collective and inclusive reflection, improving the learning processes of all actors involved and the relevance of the research focus (Cerf et al., 2012; Méndez et al., 2013; Rossing et al., 2021), as was evidenced in our study. The CRS experiment plan for the first year was modified according to new ideas from the workshops. We adjusted the CC management to prioritize weed suppression rather than nutrient supply, altered methods and timing of the CC termination, and adjusted the NEM application method. In the second year, four main modifications emerged from the workshops: increasing CC sowing density to increase soil cover and weed suppression, avoiding all synthetic pesticide applications to promote soil health and the effectiveness of the NEM application, starting NEM applications before onion transplanting, and including biofertilizer to provide N.

During this participatory research, the analytical framework was essential for a systemic shared view of the research problem among participants (Fig. 1). The understanding that crop performance or weed pressure are emergent properties of the entire cropping system and that the treatments generate long-term effects helped all participants appreciate the study's value as a step towards a more agroecological way of farming. During the project and workshops, we focused on the

processes and mechanisms underlying onion crop performance rather than testing a particular technology or individual practice (Carr et al., 2013). Quotes from participant interventions during the workshops and end-of-project interviews show the relevance of this approach: "It is all connected, if we increase the decomposition of the residues to have more nutrients, they will be gone faster, and we will have more weeds (...) we have to delay weed appearance as much as possible, for nitrogen, I think there are more tools we can use", "We still need to adjust the management better to reduce weeds, but having a good soil and adjusting the timing of fertilization a little more, the truth is that we could achieve good results with reduced tillage, we need more time in this research project (...) It will take time for the soil to respond to this good management, it is medium to long term if we have been doing everything the other way around for dozens of years, we cannot ask for miracles. We will be in a better starting position for the second year and those that follow". We conclude that the participatory approach requires a high commitment and time investment of farmers and researchers but is an effective way to develop management systems that rely on the complex interactions of agroecosystem components.

5. Conclusions

Here, we pioneered a cover crop – reduced tillage (CC-RT) system for onions without using herbicides and synthetic fertilizers in a participatory setting. This CC-RT system reduced soil erosion risk, increased biological activity, and did not pose soil physical restrictions for the crop. However, it resulted in N limitation that reduced onion yield and in high weed pressure that increased labour demand. Thus, CC-RT systems are within reach, but further research targeting effective ways to suppress weeds and increase soil N availability at the start of the growing season is needed to make them feasible for no or low agrochemical input systems. The participatory setting improved the experimental design and management. It promoted the learning processes of all actors involved, which is essential for developing long-term research platforms for agroecological farming.

Declaration of Competing Interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Mariana Scarlato reports financial support was provided by National Research and Innovation Agency of Uruguay. Mariana Scarlato reports financial support was provided by National Institute of Agricultural Research of Uruguay. Walter Rossing reports financial support was provided by WO-WOTRO.

Data Availability

Data will be made available on request.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.still.2024.106061.

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