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Effects of digestate solid fraction fertilisation on yield and soil carbon dioxide emission in a horticulture succession

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Abstract

The aim of this study was to evaluate the agronomical and environmental effects of digestate solid fraction (DSF) used as fertiliser in a vegetable crop succession (green bean, savoy cabbage, cabbage and cauliflower) in Northeast Italy (45°20' N; 11°57' E). Three fertilisation treatments were tested using DSF to substitute 0% (Tmin), 50% (T50) and 100% (T100) optimal level of mineral nitrogen fertilisation. The experiment was carried out from 22nd May 2014 (green bean sowing) to 3rd June 2015 (cabbage harvest). Summer and spring crops did not show significantly different marketable yield among fertilisation treatments with an average value (±standard error) of 9.0±0.5, 9.9±1.2 and 51.3±6.4 Mg ha^{-1} for green bean, cauliflower and cabbage, respectively. Lower DSF fertilisation effect was monitored on winter crop (savoy cabbage) with a marketable yield reduction of -35.1% than mineral fertilisation (25.9 Mg ha^{-1}), whereas the T50 treatment was not significantly different compared to the two previous ones. Crop species significantly influenced the N use efficiencies with negative recovery and use efficiency indexes for the DSF fertilisation treatments. Soil CO_2 emissions were not significantly influenced by fertilisation in all studied crops with median values always lower than 1 g m^{-2} h^{-1}.

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Introduction

The biomass anaerobic digestion for biogas production is one of the most promising renewable energy forms. Together with biogas, anaerobic digestion produces a residual material (digestate) whose adequate management or disposal must be addressed in order to avoid any constraint to the development of anaerobic digestion systems (Alburquerque *et al.,* 2012) and improve the sustainability of this renewable energy form production (Teglia *et al.,* 2011). Considering digestate characteristic chemical composition (Tambone *et al.,* 2010), biological stability (Tambone *et al.,* 2009) and the higher hygienic quality of digestate than input biomasses (Bonetta *et al.,* 2011; Goberna *et al.,* 2011) it could be used in agriculture as organic fertiliser (Cavalli *et al.,* 2014; Nkoa, 2014) with positive effects on social, productive and environmental problems. In fact, intensive agriculture has caused social and environmental problems worldwide over the few past decades and some of the most important impacts are loss of soil organic matter, soil erosion and water pollution (Zhao *et al.,* 2009). In the last years interest in the use of organic matrices in agriculture has been increasing due to the high cost of mineral fertilisers and new environmental regulations that limit their use. The recycling of organic waste materials can help maintain soil organic matter and nutrients levels, and exert positive effects on various aspects of soil fertility (Casacchia *et al.,* 2012; Marchetti *et al.,* 2012; Barbera *et al.,* 2013). On the other hand, organic matter addition can promote soil greenhouse gases emission, such as carbon dioxide (CO_2) (Li *et al.,* 2013), stimulating soil microbial activity. It is generally accepted that the quality of organic matter influences microbial activity. Particularly, the increase in soil organic matter decomposition rate after fresh organic matter input to soil, is often supposed to result from a global increase in microbial activity due to the higher availability of energy released from the decomposition of fresh organic matter (Fontaine *et al.,* 2003). In view of this, stable organic waste materials should be added to the soil to increase the soil organic matter content and to reduce soil CO_2 emission. During anaerobic digestion the easily degradable organic compounds is mineralised obtaining as output digested organic material characterised by more stable organic matter (Marcato *et al.,* 2009; Tambone *et al.,* 2009). It has been found in laboratory condition that soil microbial activity is reduced when organic residues are anaerobically digested before it distribution in the soil (Thomsen *et al.,* 2013), indicating that the organic matter in the digestate can hardly be used as carbon and energy source by soil microorganisms (Bachmann *et al.,* 2014).

A few conflicting results about the effect of digestates on crop yields have been recently reported in the literature (Möller and Müller, 2012) with results that can be grouped into three categories of performances: similar to the unfertilised control, similar or higher than undigested feedstock and equal or better than mineral fertilisers (Nkoa, 2014). Suboptimal performances of digestate involved inappropriate storage and/or application techniques that can lead to the loss of its fertiliser value or nitrogen use efficiency, through ammonia volatilisation, leaching and runoff into surface and ground waters (Nkoa, 2014). Several studies have investigated the effect of digestate fertilisation, especially on cereal and/or biomass production (Loria *et al.,* 2007; Chantigny *et al.,* 2008; Bachmann *et al.,* 2014; Maucieri *et al.,* 2016). Considering digestate effect on vegetables production literature data are also present, but several studies consider: i) one crop cycle (Hossain *et al.,* 2014); ii) the cycles of the same species in different years (Montemurro *et al.,* 2010; Lošák *et al.,* 2016); iii) different cycles of one species in the same year (Nicoletto *et al.,* 2014). Few studies consider the continuous use of digestate in a vegetable crop succession (Alburquerque *et al.,* 2012). With this in mind, the aim of this study was to evaluate the agronomical and environmental (soil CO_2 emission) effects of the digestate solid fraction (DSF) use as nitrogen source in a vegetable crop succession.

Materials and methods

Experimental description

The trial was carried out at the Experimental Farm of Padua University at Legnaro, North-East Italy (45°20' N; 11°57' E) in open field conditions using DSF on green bean (*Phaseolus vulgaris* L.,), savoy cabbage (*Brassica oleracea* var. *sabauda* L.), cabbage (*Brassica oleracea* var. *capitata*) and cauliflower (*Brassica oleracea* var. *botrytis*).

Three fertilisation treatments were tested using DSF to substitute mineral nitrogen (N) crop requirements: i) 50% N through DSF and 50% N through mineral fertiliser (T50); ii) 100% N through DSF (T100); iii) 100% mineral fertilisation (Tmin). DSF derived from a process of anaerobic digestion of cattle slurry and manure, maize silage and flour; its chemical composition is reported in Table 1.

The phosphorus (P) and potassium (K) content in the DSF were taken into consideration to calculate the amount of these elements to supply as mineral fertilisers in the different treatments (Table 2) to obtain the same amount. When P or K supplied with DSF in the T50 and/or T100 treatments were lower than the optimal dose supplied in the Tmin treatment the difference was integrated with mineral fertilisers. This fertilisation management was chosen to highlight the effects of the form in which the N was supplied.

N, P and K rates from mineral fertilisers were supplied according to standard recommendations in the area: 40, 50, 100 kg ha^{-1} for green bean and 110, 70, 160 kg ha^{-1} for the other species respectively for N, P_2O_5 and K_2O (Perelli *et al*., 2009). Both mineral and DSF were supplied from 1 to 4 days before sowing or transplanting and immediately incorporated by rotavator.

The soil was a fulvi-calcaric Cambisol with a loamy texture; its chemical characteristics, determined before the beginning of the experiment, are reported in Table 3.

A randomised block experimental design with three replications was used and plots were 40 m^2 wide (10 m × 4 m). The green bean was sowed on 22 May 2014 and harvested on 14 July 2014, savoy cabbage was transplanted on 12 August 2014 and harvested on 15 January 2015. After, each plot was split into two subplots of 20 m^2 (10 m × 2 m) and both cabbage and cauliflower were together grown transplanting them on 3 April 2015. Cauliflower was harvested on 27 May 2015 and the cabbage on 3 June 2015.

Meteorological variables

The following meteorological data were recorded by the weather station that was close to the experimental site: rain (mm), max, min and average air temperature (°C), wind speed (m s^{-1}), relative humidity (%), and solar radiation (MJ m^{-2} d^{-1}).

Plants harvest and measurements

Crops were harvested at full crop marketable maturity in a subarea of 4.5 m^2 in the inner part of each plot to determine: total biomass production, marketable yield and waste biomass. Marketable harvest index was calculated using the following equations:

Harvest index (HI) = Marketable fresh biomass/Total fresh biomass

For each plot, the marketable and waste biomass of five plants were cut into small pieces and dried in a ventilated oven at 65°C to calculate the dry matter content. In this last, the total organic nitrogen content was determined according to ISO1656 method.

Soil carbon dioxide flux measurement

CO_2 flux was measured with the static non-stationary chamber technique (Maucieri *et al.,* 2016) using a chamber with a volume of 5 L and 10 cm square base. CO_2 emissions were detected in two points of each plot in order to replicate the measures in the space with 6 measures for each studied treatment. Soil CO_2 emission was measured 13 times during green bean crop season and 12 times during the crop seasons of the other species. For savoy cabbage soil CO_2 emissions were not detected from half October till the first week of December.

Table 1. Chemical characteristics of digestate solid fraction used in the experiment.

Parameters	Values
Electrical conductivity (dS m^{-1})	1.15
pH	8.9
Dry matter (%)	22.6
Organic carbon (%)	51.5
Total organic nitrogen (%)	1.9
NH_4 (mg kg^{-1} DM)	10012.7
NO_3 (mg kg^{-1} DM)	395.7
PO_4 (mg kg^{-1} DM)	4265.9
P (mg kg^{-1} DM)	10229.6
K (mg kg^{-1} DM)	13938.9
Ca (mg kg^{-1} DM)	6295.8
Na (mg kg^{-1} DM)	2249.3
Mg (mg kg^{-1} DM)	3656.5
Cl (mg kg^{-1} DM)	5062.0
SO_4^- (mg kg^{-1} DM)	1692.7
Cd (mg kg^{-1} DM)	< 0.001
Cr (mg kg^{-1} DM)	1.0
Cu (mg kg^{-1} DM)	7.0
Hg (mg kg^{-1} DM)	< 0.001
Ni (mg kg^{-1} DM)	1.2
Pb (mg kg^{-1} DM)	0.1
Zn (mg kg^{-1} DM)	130.7

DM, dry matter.

Soil CO_2 flux was determined by measuring the temporal change in CO_2 concentration inside the chamber using a portable IR instrument (Geotech G150), detecting CO_2 concentrations at levels of parts per million. CO_2 flux was calculated using the following formula:

$$CO_2 = V/A \cdot dc/dt$$

where CO_2 flux is expressed in mg CO_2 m^{-2} s^{-1}; V (m^3) is the volume and A (m^2) the footprint of the flux chamber; c is the CO_2 concentration (mg CO_2 m^{-3}) and t the time step (s).

In each CO_2 measurement point, soil temperature and moisture (TDR 100 Soil Moisture Meter) in the first 7.5 cm were also detected.

Nitrogen use efficiency

N harvest index was calculated using the following equations:

N harvest index (NHI) = N uptake in marketable dry biomass/N uptake in total dry biomass

N use efficiency (NUE) was evaluated using the approach suggested by Fageria *et al.* (2010) calculating: agronomic efficiency (AE), physiological efficiency (PE), agrophysiological efficiency (APE), apparent recovery efficiency (ARE), utilisation efficiency (EU). Nitrogen indexes were calculated using the following equations:

$$AE\ (mg\ mg^{-1}) = (Gf–Gu)/Na$$
$$PE\ (mg\ mg^{-1}) = (BYf–BYu)/(Nf–Nu)$$
$$APE\ (mg\ mg^{-1}) = (Gf–Gu)/(Nf–Nu)$$
$$ARE\ (\%) = [(Nf–Nu)/Na] \times 100$$
$$EU\ (mg\ mg^{-1}) = PE \times ARE$$

where Gf is the marketable yield of the DSF fertilised plots (mg) (T50 or T100), Gu is the marketable yield of the mineral fertilised plots (mg) (Tmin), and Na is the quantity of nitrogen applied (mg), BYf is the biological yield (total biomass) of the DSF fertilised plots (mg) (T50 or T100), BYu is the biological yield of the mineral fertilised plots (mg) (Tmin), Nf is the nitrogen uptake (total biomass) of the DSF fertilised plots (T50 or T100), and Nu is the nitrogen uptake (total biomass) of the mineral fertilised plots (mg) (Tmin).

Digestate solid fraction analysis

The pH and electrical conductivity of DSF were determined according to EN13037 and EN13038, respectively. Dry matter was calculated following EN13040 and organic matter using EN13039. organic nitrogen was measured according to ISO1656; total organic carbon was calculated according to Nelson *et al*. (1996). Total contents of P, K, Ca, Mg, Cd, Cr, Cu, Ni, Pb, Zn, Hg and Na were determined using inductively coupled plasma atomic-emission spectrometry (ICP-AES) SPECTRO Ciros (Spectrum Italy Srl, Basiglio, Italy). In addition to fully characterise DSF, NH_4, NO_3, PO_4, Cl and SO_4 were determined using an ion chromatography system (ICS-900, Dionex Corp.) These analyses were conducted on DSF ash (Zancan *et al.,* 2006) and in water extracts (1:6, v/v) in order to check the soluble amount of macro and micro-nutrients.

Carbon dioxide equivalent balance

Considering only the DSF macronutrients (N, P_2O_5, K_2O), and using the CO_2 equivalent ($CO_{2(eq)}$) specific emission factors for mineral fertilisers production the environmental impact of fertilisation was analysed. The specific emission factors adopted to estimate the $CO_{2(eq)}$ balance were 3.26 kg $CO_{2(eq)}$ emitted for each kg of N, 2.01 kg $CO_{2(eq)}$ emitted for each kg of P_2O_5 and 1.41 kg $CO_{2(eq)}$ emitted for each kg of K_2O (Capponi *et al.,* 2012). The $CO_{2(eq)}$ emissions due to the nutrients supplied by mineral fertilisers were counted as positive emission whereas the $CO_{2(eq)}$ emissions due to the nutrients supplied by DSF were counted as avoided emissions with negative sign. The $CO_{2(eq)}$ balance was calculated in terms of $CO_{2(eq)}$ ha^{-1}.

Statistical analysis

Bio-agronomics data were analysed using ANOVA and the comparison between means was made using Fisher LSD test. NUE means were statistically processed by Fisher LSD test to compare crop species and by t student test to compare fertilisation treatments. Soil CO_2 emission data were not normal distributed therefore they were analysed with Kruskal-Wallis nonparametric test. Correlation between soil temperature and moisture with CO_2 emissions were evaluated using Spearman Rank correlation.

Table 2. Macronutrients applied as mineral fertilisers or digestate solid fraction during horticulture succession.

	Mineral fertilisers (kg ha^{-1})			Digestate (kg ha^{-1})		
	N	P_2O_5	K_2O	N	P_2O_5	K_2O
Green bean						
Tmin	40.0	108.7	212.7	0	0	0
T50	20.0	34.0	172.9	20.0	74.7	39.8
T100	0	0	133.0	40.0	149.4	79.7
Savoy cabbage						
Tmin	110.0	152.2	340.4	0	0	0
T50	55.0	0	230.7	55.0	205.3	109.7
T100	0	0	121.1	110.0	410.6	219.3
Cabbage						
Tmin	110.0	152.2	533.3	0	0	0
T50	55.0	0	378.0	55.0	161.8	155.3
T100	0	0	222.6	110.0	323.6	310.6
Cauliflower						
Tmin	110.0	152.2	533.3	0	0	0
T50	55.0	0	378.0	55.0	161.8	155.3
T100	0	0	222.6	110.0	323.6	310.6

Tmin, 100% of nitrogen crop requirement satisfied through mineral fertilisation; T50, 50% of nitrogen crop requirement satisfied through mineral fertilisation and 50% throught digestate solid fraction; T100, 100% of nitrogen crop requirement satisfied through digestate solid fraction.

Results and discussion

Meteorological variables

Meteorological data recorded during the experimental period are reported in Figure 1. Cumulative rainfall was 1016.4 mm (208.4 mm during green bean cultivation, 414.8 mm during savoy cabbage and 198.4 mm during cabbage and cauliflower cultivations), 16% higher than the average rain monitored in the same period in the previous 20 years. The average daily air temperature was 14.4°C with its maximum value at the end of the green bean crop season (June 12th 2014, 34.0°C) and minimum value after savoy cabbage harvest (January 28th 2015, -4.4°C). The average solar radiation was 13.7 MJ m^{-2} d^{-1}, with the higher monthly average intensity value registered in June 2014 (23.8 MJ m^{-2} d^{-1}); average daily wind speed was 2.0 m s^{-1}.

Plants harvest and measurements

The crops yield was not significantly influenced by the fertilisation treatments with an average (±standard error) marketable yield of 9.0±0.5, 9.9±1.2 and 51.3±6.4 Mg ha^{-1} for green bean, cauliflower and cabbage, respectively. The savoy cabbage yield was significantly higher in the Tmin treatment than T100 one; T50 was not significantly different between the previous two treatments considering marketable yield whereas significant higher production was detected considering no marketable and total biomass than T100 treatment (Table 4).

Considering savoy cabbage yield, similar trend among fertilisation treatments was previously reported in the same areas by Nicoletto *et al.* (2012), although with higher absolute yield values. The different production is due to the lower N fertilisation in this study (-26.7%) than that supplied by Nicoletto *et al.* (2012) that determine a marketable yield reduction of 26.1, 38.8 and 37.8% in the Tmin, T50 and T100 treatments, respectively. The higher per-

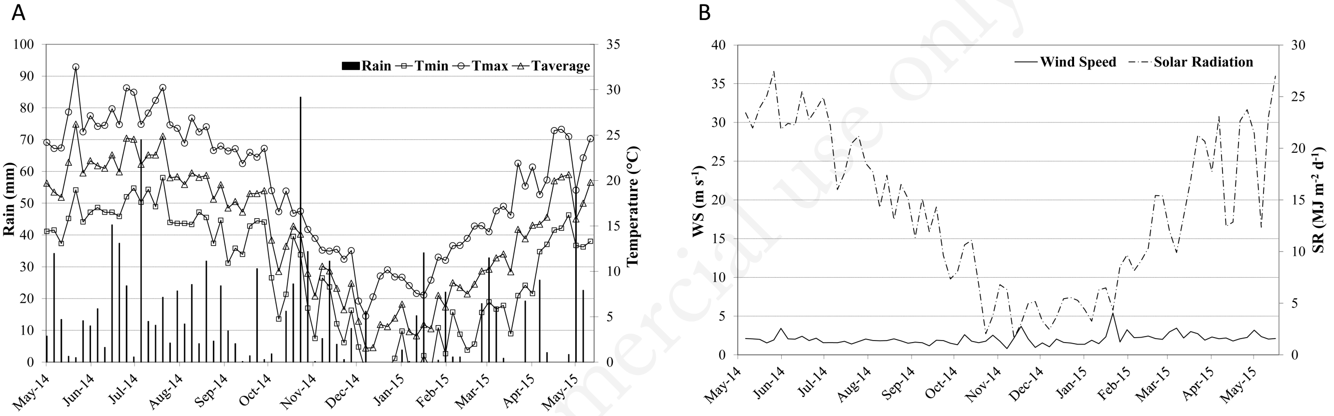

Figure 1. Meteorological data during experimental period. Five-day averages for temperatures and five-day cumulative rainfall (A), wind speed and solar radiation (B).

Table 3. Soil chemical properties on dry matter basis.

Parameters	Soil depth (m) 0.00-0.30	0.30-0.60
Nitrogen (%)	0.11	0.10
Organic carbon (%)	0.87	0.78
pH	7.57	7.77
Electrical conductivity (dS m^{-1})	0.27	0.25
Nitrites (mg kg^{-1})	3.16	4.09
Sodium (mg kg^{-1})	22.4	18.0
Nitrates (mg kg^{-1})	23.8	20.0
Phosphates (mg kg^{-1})	4.43	2.89
Sulphates (mg kg^{-1})	59.9	23.9
Magnesium (mg kg^{-1})	32.2	29.6
Calcium (mg kg^{-1})	256	239
Chlorides (mg kg^{-1})	13.4	4.46
Potassium (mg kg^{-1})	14.3	12.0

Table 4. Fresh biomass production (Mg ha^{-1}).

	Marketable biomass	No marketable biomass	Total biomass
Green bean			
Tmin	8.15±0.62	11.50±0.33	19.65±0.79
T50	9.36±0.86	11.98±0.94	21.34±1.74
T100	9.50±0.90	11.84±1.04	21.34±1.94
Savoy cabbage			
Tmin	25.87±1.03^{a}	25.58±1.79^{a}	51.44±2.76^{a}
T50	20.81±2.33^{ab}	26.91±2.93^{a}	47.72±1.91^{a}
T100	16.80±2.67^{b}	16.12±1.85^{b}	32.92±4.52^{b}
Cabbage			
Tmin	65.97±11.41	33.61±9.82	99.59±20.93
T50	41.14±7.15	25.42±0.77	66.56±7.91
T100	46.79±11.59	27.96±4.80	74.74±16.33
Cauliflower			
Tmin	10.67±2.42	23.38±2.45	34.04±4.86
T50	8.94±2.95	24.88±3.52	33.82±4.13
T100	10.09±1.74	19.29±0.34	29.38±2.06

Tmin, 100% of nitrogen crop requirement satisfied through mineral fertilisation; T50, 50% of nitrogen crop requirement satisfied through mineral fertilisation and 50% throught digestate solid fraction; T100, 100% of nitrogen crop requirement satisfied through digestate solid fraction. ^{a,b}Different letters indicate significant differences for Fisher Least Significant Difference test at $P<0.05$.

centage reduction in our treatments with DSF respect Nicoletto *et al.* (2012) is maybe due to the different experimental period. In fact, in our study savoy cabbage was transplanted about four months after experiment beginning whereas in Nicoletto *et al.* (2012) after 2 years with higher nutrients (especially N) availability due to the residual effect of the previous crops cycles; moreover different cultivar were used. However, the one year succession results showed comparable fertilisation effect of digestate respect mineral fertilisation on summer and spring crops at both quantities supplied (T50 and T100) whereas lower DSF fertilisation effect was monitored for winter crop when N fertilisation was carried out using only digestate (T100). The lower production with DSF was probably due to the low environmental temperatures that reduced soil microbial activity, with a consequent reduction of the digestate nutrients availability in agreement with Alburquerque *et al.* (2012) and our previous results (Nicoletto *et al.,* 2014). Considering that the yield of spring and summer crops is not influenced by the partial or total replace of inorganic fertilisers, this suggested that the use of digestate may reduce agricultural dependence on mineral fertilisers and the energy and economic costs associated with their use (Walsh *et al.,* 2012).

As expected significant variations of HI and NHI were observed among crops. No significant differences were found between green bean and savoy cabbage with an average HI and NHI of 0.46 and 0.39.

Crop species, on the average of DSF fertilisations (T50 and T100), significantly influenced the AE, ARE and UE indexes (Table 5), with negative indexes values for all species except for green bean, which showed positive values probably due to the higher symbiotic N_2 fixation activity in the DSF treatments than 100% mineral one. Considering the AE, although cabbage and cauliflower were cultivated in the same period, they showed significant different values with lower AE cultivating cabbage. This difference can be traced at the different marketable part of the two species, vegetative part (leaves) in cabbage and reproductive part (inflorescence) in cauliflower. As well known, plants have higher N requirement to produce vegetative part than reproductive one. Considering the short DSF application time we can assume that DSF was not able to satisfy the cabbage N requirement. In fact, in the same year of the experiment reported in this paper, significant different AE values comparing cabbage and cauliflower cultivated in a long term DSF fertilisation trial (10 years) (unpublished data) were not observed. In view of this, to maximise the AE of N supplied with DSF, our results suggest that in the short-term (one year succession) cauliflower should be preferred at cabbage as spring crop.

Fertilisation treatment, on the average of crop species, exerted a significant effect only on ARE with negative values in both treatments with DSF (T50 and T100) and lower N recovery efficiency in the T100 than T50 (Table 6).

The negative ARE and UE indexes can be traced at the lower N availability in DSF treatments because in these last the N was supplied mainly in organic form and so need mineralisation process before crops uptake. Considering the slow turnover of organic matter in the soil, long-term studies are necessary to evaluate the nutrients use efficiency in agro-ecosystems managed for several years with DSF.

In economic terms the use of DSF has further advantages and some considerations have to be taken into account about the potential savings obtained by the farmer. Currently the cost of anaerobic digestate residues for fertilisation purpose can be quantified with only the transport and distribution of the raw material amounting to around 1.77 € for N units supplied with DSF. The same units of N provided with urea costs about 2.30 € (fertiliser purchase and distribution); consequently the application of N through digestate leads to an economic saving of about 23% in the fertilisation costs. Moreover, the addition of organic matter, together with the macronutrients required by the crop, involves considerable agronomic advantages in the long period by increasing the soil fertility. Additionally, the possibility of storing in the soil considerable amount of organic carbon, is aligned with what is required by recent European regulations relating to carbon management and the closure of the waste recovery cycle (Arthurson, 2009).

Soil carbon dioxide flux measurement

Soil CO_2 emissions were not significantly influenced by fertilisation in all studied crops with median values always lower than 1 g m^{-2} h^{-1}. Our results are in agree with Alburquerque *et al.* (2012) who reported no significant differences in terms of soil CO_2

Table 5. Nitrogen use efficiency of four studied species on the average of digestate solid fraction fertilisation (T50 and T100).

Species	AE (mg mg^{-1})	PE (mg mg^{-1})	APE (mg mg^{-1})	ARE (%)	UE (mg mg^{-1})
Green bean	2.71^{a}	27.46^{a}	0.44^{a}	18.16^{a}	6.46^{a}
Savoy cabbage	-4.75^{ab}	24.85^{a}	1.33^{a}	-19.39^{b}	-5.51^{ab}
Cabbage	-11.21^{b}	18.12^{a}	14.39^{a}	-52.66^{b}	-14.68^{b}
Cauliflower	-0.93^{a}	19.94^{a}	-9.11^{a}	-17.27^{ab}	-1.95^{ab}

AE, agronomic efficiency; PE, physiological efficiency; APE, agrophysiological efficiency; ARE, apparent recovery efficiency; UE, utilisation efficiency. ^{a,b}Different letters indicate significant differences for Fisher Least Significant Difference test at P<0.05.

Table 6. Nitrogen use efficiency of two digestate treatments on the average of crop species.

Treatment	AE (mg mg^{-1})	PE (mg mg^{-1})	APE (mg mg^{-1})	ARE (%)	UE (mg mg^{-1})
T50	-3.79^{a}	29.74^{a}	3.72^{a}	-5.15^{a}	-2.06^{a}
T100	-3.16^{a}	14.59^{a}	-0.33^{a}	-31.43^{b}	-5.80^{a}

AE, agronomic efficiency; PE, physiological efficiency; APE, agrophysiological efficiency; T50, 50% of nitrogen crop requirement satisfied through mineral fertilisation and 50% throught digestate solid fraction; T100, 100% of nitrogen crop requirement satisfied through digestate solid fraction. ARE, apparent recovery efficiency; UE, utilisation efficiency. ^{a,b}Different letters indicate significant differences for t-student test at P<0.05.

emissions between mineral and digestate fertilisation in watermelon-cauliflower succession. The no significant different soil CO_2 emission among fertilisation treatments (Tmin, T50, and T100) can be ascribed at the characteristics of organic matter content in the DSF. In fact, fresh organic matter addition can promote soil CO_2 emission due to its easily decomposition by microbial activity (Fontaine *et al*., 2003), in opposite stabilised organic matter can maintain stable or decrease soil CO_2 emission reducing microbial activity (Thomsen *et al*., 2013; Bachmann *et al*., 2014). The DSF used in this experiment came from a mesophilic (35-40°C) anaerobic digestion plant that had a substrate retention time of 88-92 days. Therefore, due to anaerobic digestion process characteristics, we can assume that with DSF was supplied stabilised organic matter that did not influence soil respiration in the experimental period (1 year). In fact, considering the N supplied (260 kg ha^{-1}) and the DSF composition (Table 1), during all succession the C added was 698 and 349 g m^{-2} in the T100 and T50 plots, respectively, whereas the C emitted from soil as total respiration was +59 and -13 g m^{-2} in T100 and T50, respectively, compared with Tmin treatment.

The green bean, cultivated from May to July showed emission peaks that reach values of 4.5 g CO_2 m^{-2} h^{-1} and an emission median value of 0.46 g CO_2 m^{-2} h^{-1}. During monitoring period soil CO_2 emissions were positively correlated ($P<0.01$) with soil moisture that ranged between 12.9 and 35.5%. No correlation was found between soil CO_2 emissions and soil temperature that ranged between 17.2 and 34.7°C.

The savoy cabbage, fall-winter crop, showed an emission median value among fertilisation treatments of 0.74 g CO_2 m^{-2} h^{-1}. In opposite of green bean, during savoy cabbage cropping season the soil CO_2 emissions were positively correlated ($P<0.01$) with soil temperature that ranged between 4.3 and 31.6°C, whereas no correlation was found with soil moisture ranged from 13.9 to 52.9%. For cabbage and cauliflower, spring crops, were monitored soil CO_2 emission median values of 0.84 g m^{-2} h^{-1} and 0.78 g m^{-2} h^{-1}, respectively. During monitoring period soil CO_2 emissions were positively correlated ($P<0.01$) with temperature (ranged from 15.3 to 31.2°C and from 11.7 to 31.5°C for cabbage and cauliflower, respectively) and negatively correlated ($P<0.001$) with soil moisture (ranged from 15.8 to 54.6% and from 15.8 to 47.7% for cabbage and cauliflower, respectively).

The seasonal variation in soil CO_2 emission is commonly attributed to change in soil temperature, moisture or both (Davidson *et al.,* 1998; Ding *et al.,* 2007, 2010; Li *et al.,* 2013). In our experiment summer and winter crops showed an opposite response considering these abiotic factors. When the minimum soil temperature is high (17.2°C in summer crop) the limiting factor is water availability for microorganism's activity whereas, during fall-winter crop season with lower temperature and wet condition soil temperature was the limiting factor. Considering spring crops, with intermediate condition than green bean and savoy cabbage, we assume that soil CO_2 emission was positively correlate with soil temperature because of the positive effect on microbial activity after winter season whereas it was negatively correlate with soil moisture because high value of this last determine poor gas diffusion in the soil (Rochette *et al.,* 1991).

CO_2 green bean emission peaks, higher than other species, should be traced at the higher soil microbial activity due to the higher soil temperature with good soil moisture content whereas the increase in the soil CO_2 emission median values following the crops cycles is probably explained by the organic matter accumulation.

The environmental impact, in terms of greenhouse gas emissions, is an important parameter to evaluate the possible substitution of mineral fertilisation with DSF in horticulture succession. Considering the $CO_{2(eq)}$ balance between the quantity emitted in the atmosphere for chemical fertilisers production and the indirect $CO_{2(eq)}$ saving due to fertilisation with DSF, in our experiment, as expected, positive emission balance was observed in the Tmin treatment. Always negative balance was calculated for T100 treatment whereas for T50 treatment it was positive for green bean crop and negative for other ones (Table 7).

Conclusions

Marketable yield of spring and summer species (green bean, cauliflower and cabbage) was not significantly influenced by fer-

Table 7. Carbon dioxide equivalent emissions to produce chemical fertilisers and saving carbon dioxide equivalent due to solid digestate use in this experiment.

	Mineral fertilisers $CO_{2(eq)}$ (kg ha^{-1}) N	Mineral fertilisers $CO_{2(eq)}$ (kg ha^{-1}) P_2O_5	Mineral fertilisers $CO_{2(eq)}$ (kg ha^{-1}) K_2O	Digestate $CO_{2(eq)}$ (kg ha^{-1}) N	Digestate $CO_{2(eq)}$ (kg ha^{-1}) P_2O_5	Digestate $CO_{2(eq)}$ (kg ha^{-1}) K_2O	Total $CO_{2(eq}$ (kg ha^{-1})
Green bean							
Tmin	130.4	100.5	141.0	0.0	0.0	0.0	371.9
T50	65.2	31.6	114.6	-65.2	-68.9	-26.4	50.9
T100	0.0	0.0	88.1	-130.4	-138.1	-52.9	-233.2
Savoy cabbage							
Tmin	358.6	140.7	225.6	0.0	0.0	0.0	724.9
T50	179.3	0.0	153.0	-179.3	-189.7	-72.6	-109.4
T100	0.0	0.0	80.2	-358.6	-379.7	-145.4	-803.4
Cabbage							
Tmin	358.6	140.7	225.6	0.0	0.0	0.0	724.9
T50	179.3	0.0	159.9	-179.3	-149.5	-65.7	-55.4
T100	0.0	0.0	94.2	-358.6	-299.1	-131.4	-694.9
Cauliflower							
Tmin	358.6	140.7	225.6	0.0	0.0	0.0	724.9
T50	179.3	0.0	159.9	-179.3	-149.5	-65.7	-55.4
T100	0.0	0.0	94.2	-358.6	-299.1	-131.4	-694.9

Tmin, 100% of nitrogen crop requirement satisfied through mineral fertilisation; T50, 50% of nitrogen crop requirement satisfied through mineral fertilisation and 50% throught digestate solid fraction; T100, 100% of nitrogen crop requirement satisfied through digestate solid fraction. Positive values indicate net $CO_{2(eq)}$ emission whereas negative values indicate avoided $CO_{2(eq)}$ emissions.

tilisation treatment. Instead, the marketable yield of winter crop (savoy cabbage) was significantly higher with N mineral fertilisation than DSF one. On the contrary, when using both N mineral fertiliser and DSF at 50% the yield was not significantly different with respect to the previous two treatments. Therefore, our results suggest that DSF can be used to substitute N fertilisation in spring and summer species whereas during winter season it should be used to integrate N fertilisation. Partial or total fertilisation using DSF determined negative values of N apparent recovery efficiency and N use efficiency due to the slow turnover of organic matter in the soil suggesting that long term studies are desirable to evaluate the nutrients use efficiency in agro-ecosystems fertilised with organic matrices. The application of N through DSF leads to an economic saving of about 23% in the fertilisation costs respect urea. Soil CO_2 emissions were not significantly influenced by fertilisation in all studied crops with median values always lower than 1 $g\ m^{-2}\ h^{-1}$. Considering the $CO_{2(eq)}$ quantity emitted in the atmosphere for chemical fertilisers production the partial or total fertilisation with DSF determined an indirect $CO_{2(eq)}$ saving.

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