



Use of field diagnostic tools for top dressing nitrogen recommendation when organic manures are applied in humid Mediterranean conditions

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Abstract. Nitrogen is often applied in excessive quantities, causing nitrogen losses. In recent years, the management of large quantities of manure and slurry compounds has become a challenge. The aim of this study was to assess the usefulness of the proxy tools Yara N-testerTM and RapidScan CS-45 for diagnosing the N nutritional status of wheat crops when farmyard manures were applied. Our second objective was to start designing a N fertilization strategy based on these measurements. To achieve these objectives, three annual field trials were established at three close locations with similar soils. Three kinds of initial fertilizers [dairy slurry (40 t ha⁻¹), sheep manure (40 t ha⁻¹) and conventional (no organic fertilizer on basal dressing and 40 kg N ha⁻¹ at tillering)] were added and five N mineral fertilization dose applied at stem elongation for each kind of initial fertilization. The proxy tools were used at stem elongation (GS-30) before applying the mineral N. Proxy tool readings look promising in order to adjust the N application rate at stem elongation. In dairy slurry, when both proxy tool values were 60 - 65 % the optimum N rate at stem elongation for achieving the maximum yield was 118 - 128 kg N ha⁻¹. In dairy slurry and conventional treatments when readings were 85 - 90 % the optimum N rate was 100 - 110 kg N ha⁻¹. However, at the sheep manure treatment, it is more difficult to find a relationship between sensor readings and yield. Moreover, when organic fertilizers were applied before sowing, the usual first N topdressing at the beginning of tillering could be avoided in years with humid spring.

Keywords. Winter wheat, Precision N fertilization, Chlorophyll meter, Canopy reflectance sensing, N Mineralization, Organic amendments

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Introduction

Few agroecosystems supply enough nitrogen (N) to sustain satisfactory crop production without fertilizers. In cereal cropping systems, N is the most critical growth restricting nutrient (Samborski et al. 2009), but to ensure that the potential yield is reached each year, N is often applied in excessive quantities causing nitrogen losses. The manufacture of mineral fertilisers requires energy, therefore recycling of nutrients throughout organic materials and improving nutrient availability from well-structured biologically active soils makes better use of resources and economic sense (Defra 2017). In recent decades, the interest in maintaining soil and water quality for a sustainable environment has increased. Moreover, with the increase in the scale of livestock farming, the disposal and management of large quantities of manure and slurry compounds pose serious environmental challenges.

N organic waste application generally has a lower availability to crops than N in mineral fertilisers (Defra 2017). It is difficult to predict the amount of available N from organic manures for plants because is influenced by the composition of organic residues, drying and rewetting events and soil characteristics (Cabrera et al. 2005). On the other hand, climate varies considerably from year to year and site to site causing large differences in yield potential; consequently, crop N fertilizer demand widely varies. Therefore, application of organic fertilisers often is combined with the application of mineral fertilisers that rapidly enter in the plant available N pool. Combining organic amendment applications with nitrogen mineral fertilizer applications, with the aim to meet crop N needs, can be a suitable alternative for the management of manures. Attaining profitable yields without increasing N losses requires an adequate synchronicity between crop N demand and N supply from all sources throughout the growing season (Shanahan et al. 2008; Samborski et al. 2009).

In the area where this study was carried out (Basque Country, northern Spain), organic compounds are usually applied as a basal dressing before sowing due to the humid climate in winter and spring that usually hinder the entrance of machinery to fields. Therefore, it is necessary to know crop N status at the time of the last and greater mineral N dressing application at stem elongation (GS-30, Zadoks et al. 1974). Real-time and accurate detection of crop N concentration and uptake is important for optimizing N management for obtaining in-season recommendations for N rate instead of applying the same N rate every year. Efforts have focused on strategies based on in-season N application, taking soil and crop N status during the vegetative period into account.

In Western Europe, the N_{min} method for N fertilization application has been widely used in cereals. The method is based on the measured amount of soil mineral nitrogen in the main rooting depth before N fertilizer application at the beginning of the rapid period of crop growth. The recommended rate of N fertilizer was calculated by the predicted N demand for the target yield minus the measured soil N_{min} value. However, the determination of N_{min} requires time-consuming procedures and translating few values of soil samplings to a heterogeneous field makes it imprecise.

In recent years, significant progress has been observed in the development and investigation of optical sensing tools used for N status determination (Samborski et al. 2009). Hand-held chlorophyll meters provide instant and non-destructive readings for plant leaves; Yara N-tester™ (Yara International ASA, Oslo, Norway) measures the light transmittance of the leaf at red (650 nm, around the maximum chlorophyll absorption) and near-infrared (NIR, 960 nm) wavelengths and calculates a numeric, dimensionless value that is proportional to the amount of total chlorophyll present in the leaf. Chlorophyll meter readings have widely proven to be well correlated with leaf chlorophyll and N concentrations in wheat; therefore, chlorophyll content can be used to diagnose the nitrogen status of plants (Ortuzar-Iragorri et al. 2005; Arregui et al. 2006) and could be used to decide if a supplementary dose is to be applied in order to increase grain N content (Arregui et al. 2006 ; Ortuzar-Iragorri et al. 2017).

Remote sensing is another option for obtaining information on crop N status for portions or the entire field providing a rapid estimate of the crop N status (Samborski et al. 2009). Ground-based crop canopy reflectance sensing has been used to determine crop N conditions and N input recommendation in cereals (Shanahan et al. 2008) and is not affected by clouds, unlike aerial or satellite sensing. For practical on-farm management, active crop canopy sensors with their own light sources are needed, so they will not be limited by environmental light conditions. Spectral data collected by the sensor can then be converted into measurements of canopy green area by calculating vegetation indices such as Normalized Difference Vegetation Index (NDVI). RapidScan CS-45 (Holland Scientific, Lincoln, NE) is an entirely self-contained ground-based active canopy sensor that measures crop reflectance at 670, 730 and 780 nm, and provides the NDVI.

The aim of this study was to assess the usefulness of the proxy tools Yara N-tester™ and RapidScan CS-45 for adjusting the optimum N rate at stem elongation (GS-30) when farmyard manures are applied before sowing in humid Mediterranean conditions.

Material and methods

Three field trials were established in Arkaute (Araba, Basque Country, Spain) at NEIKER-Tecnalia facilities (42850N, 2620W; elevation 515 m above sea level) in three consecutive wheat growing seasons 2014–2015, 2015–2016 and 2016–2017 (defined as 2015, 2016 and 2017) in different fields under rainfed conditions. There was 130 m distance among the three field trials. In the field trial used in 2015, a soil pit was made, and after describing and analysing its horizons, the soil was classified as Hypercalcic Kastanozem (IUSS 2015). Mineralogical properties of the soil were analysed (Analysis by X-ray diffraction); Soil presented 40 % of clays (Clays < 2 µm: Illite (%) = 30; Kaolinite (%) = 12; Smectite (%) = 58). In field trials of 2016 and 2017, several prospective holes were observed, verifying that the three soils had similar characteristics. The three fields were flat.

Representative soil samples were taken from each field trial for analysing the physical and chemical properties before wheat sowing from depths 0–30 and 30–60 cm. Soil texture was analysed by pipette method (Gee and Bauder 1986) and classified (USDA 1999; 0–30 cm, sandy clay loam and 30–60cm, clay loam). Soils had high pH values (1:2.5 soil:water using a pH-Meter CG840; 8.0–8.5), were calcareous (MAPA 1994; 3.6%-58% according to soil depth) and had moderate organic matter content (Walkey and Black 1934) in the upper layer (2–2.5 %).

Experimental setup and treatments

Three kinds of initial fertilization were applied: dairy slurry (40 t ha⁻¹), sheep farmyard manure (40 t ha⁻¹) and conventional (no organic fertilizer basal dressing and 40 kg N ha⁻¹ at tillering). These three types of fertilization were combined with five N rates (calcium-ammonium-nitrate, NAC 27 %) at top dressing applied at stem elongation (0, 40, 80, 120 and 160 kg N ha⁻¹). Apart from the treatments, two controls were established: a control without N fertilization (0 N) and an overfertilized control plot (280; 80 kg N ha⁻¹ applied at tillering and 200 kg N ha⁻¹ applied at stem elongation; Table 1). The experiment was a factorial randomized complete block design with three factors (year, initial fertilization and N rate at stem elongation) and four replicates. The gross area of each plot was 4 m wide and 8 m long.

Organic amendments were applied on 13/11/2014, 4/11/2015 and 17/11/2016. Slurry and manure were sampled and analysed for total N and NH₄⁺-N (Table 1). We decided to apply manure at 40 t ha⁻¹ and slurry at 40 t ha⁻¹ because this is the usual rate when organic amendments are applied as initial fertilizers in Araba, Basque Country, Spain. Commonly, organic manures are rotationally applied every two or three years in cereal crops. Conversely, when organics are not applied as initial fertilizers, the usual agricultural practice in Araba is to apply 40 kg N ha⁻¹ at tillering, as we did at conventional treatments (Table 1).

Wheat (*Triticum aestivum* var. Cezanne) was sown on 24/11/2014, 06/11/2015 and 18/11/2016, and was harvested on 21/07/2015, 2/08/2016 and 2/08/2017. The sowing rate was 220 kg seed ha⁻¹. The preceding crops were flax (*Linum usitatissimum*), rapeseed (*Brassica napus*) and wheat (*Triticum aestivum*), respectively.

Mineral N samples (N_{min})

Three soil samples from replicates I, II and III were taken for each kind of initial fertilization treatment (dairy slurry, sheep farmyard manure and conventional), exactly in 40N+0N, DS+0N and SM+0N treatments (Table 1) at 0–30 in two moments. 1) at the end of winter (09/03/2015, 19/01/2016 and 02/03/2017) just before mineral N fertilization (GS-21 beginning of tillering; Zadoks et al. 1974) at the conventional treatment. 2) at the beginning of stem elongation (GS-30; 04/04/2015, 17/03/2016 and 06/04/2017) just before mineral N fertilization at (GS-30).

The samples were analysed for soil mineral nitrogen ($\text{NH}_4^+\text{-N}$ plus $\text{NO}_3^-\text{-N}$) by spectrophotometry (Cawse 1967; Nelson 1983; Wei et al. 2008).

Table 1. N application rates and timing for three initial fertilization treatment for field trials (2015, 2016 and 2017) and control (0N) and overfertilized (280N) plots.

Initial fertilization	2015		2016		2017		2015-2016-2017		Treatment identification
	Total N ^a (kg ha ⁻¹)	N-NH ₄ ^{+b} (kg ha ⁻¹)	Total N ^a (kg ha ⁻¹)	N-NH ₄ ^{+b} (kg ha ⁻¹)	Total N ^a (kg ha ⁻¹)	N-NH ₄ ^{+b} (kg ha ⁻¹)	Topdressing at tillering (kg N ha ⁻¹)	Topdressing at stem elongation (kg N ha ⁻¹)	
Conventional (--)	--	--	--	--	--	--	40	0	40N+0N
								40	40N+40N
								80	40N+80N
								120	40N+120N
								160	40N+160N
Dairy Slurry (40 t ha ⁻¹)	192	104	144	80	120	68	--	0	DS+0N
								40	DS+40N
								80	DS+80N
								120	DS+120N
								160	DS+160N
Sheep manure (40 t ha ⁻¹)	336	0	592	200	448	--	--	0	SM+0N
								40	SM+40N
								80	SM+80N
								120	SM+120N
								160	SM+120N
Control	--	--	--	--	--	--	--	--	0N
Overfertilized	--	--	--	--	--	--	80	200	280N

^aTotal N (dry combustion using a LECO TruSpec®CHNs, ^bKjeldahl digestion(MAFF, 1986)

Proxy tools for adjusting the optimum N rate at stem elongation (GS-30)

The proxy tools for the diagnosis of the N nutritional status tested were Yara N-tester™ (Yara International ASA, Oslo, Norway) and RapidScan CS-45 (Holland Scientific, Lincoln, NE).

Measurements were taken in four replications in each kind of initial fertilization treatment (dairy slurry, sheep farmyard manure and conventional) just before applying the top dressing at the beginning of stem elongation (GS-30). Measurements were taken exactly in 40N+0N, DS+0N and SM+0N treatments (Table 1). In addition, samples were taken at control (0N) and overfertilized treatments (280N).

The measurements with Yara N-tester™ and RapidScan CS-45 were taken as described at Aranguren et al. (2017). Normalized values for Yara N-tester™ and RapidScan CS-45 measures were calculated to avoid the noise encountered by variables other than N fertilizer. These values were calculated as a percentage by assigning the 100% value to the overfertilized treatment (280 N) described before, similar to the technique suggested by Follett and Follett (1992).

Grain yield

Yields were recorded at crop maturity using a plot harvester (1.5 m x 8 m). For comparisons among fields, yields were converted to 12% dry matter basis.

To show grain yield response to N fertilization at stem elongation (GS-30), the grain yield was determined by fitting a Quadratic Plateau Function because it has been shown to best describe the yield response to N fertilization in humid Mediterranean climate conditions of Araba (Basque Country, northern Spain) after comparison with other models (Ortizar-Iragorri et al. 2010). A Quadratic Plateau Function was used to indicate the optimum N rate at stem elongation to achieve the maximum yield (yield vs. fertilizer N rate at stem elongation). The most important attribute of the function is where yield becomes relatively insensitive to increases in N fertilizer addition at stem elongation.

The equations obtained with the Quadratic Plateau Function were used to determine the economically optimal dose of N, following the technique suggested by Aizpurua et al. (2010). According to their findings, the revenues obtained can be calculated as:

$$\text{Revenue yield} = wY - fN$$

where w is the wheat price (€ kg⁻¹), f is the fertilizer price (€ kg⁻¹), N is the nitrogen rate (kg ha⁻¹) and Y is the quadratic plateau function.

When the revenue yield is derived respect to N rate and equalized to zero, maximum revenues would be obtained, and that would be the economical optimum rate based on yield. The wheat grain price used was 0.18 € kg⁻¹, and the fertilizer price was 1.19€ kg N (MAPAMA 2017).

Statistical analysis

Soil mineral nitrogen and proxy tool (Yara N-testerTM and RapidScan CS-45) measurements were conducted before applying the N fertilization rate at stem elongation. Therefore, the factors analysed for statistical analysis were growing season and initial fertilization by analyses of variance (ANOVA) using 'R 3.2.5' software (R core Team 2013). To separate the means, Duncan's test was used ($p \leq 0.05$) using the *R package agricolae* (De Mendiburu 2009). Overfertilized plots were not included in the ANOVA. In the case of Yara N-testerTM and RapidScan CS-45 the overfertilized plots were used to relativize the values. There was an interaction between growing season and initial fertilization for all these measurements. Therefore, an ANOVA was performed to analyse differences among initial fertilization treatments in each growing season. Another ANOVA was performed to analyse the differences among growing seasons in each initial treatment. The Quadratic Plateau Function was used for the wheat yield response. A nonlinear regression procedure was carried out using *R 3.2.5' software* (R core Team 2013) to plot curves that best described the yield response to N fertilizer application.

Results and Discussion

Grain yield

The optimum N rate at stem elongation was different for each kind of initial fertilizer (conventional, slurry or sheep manure) in each wheat growing season. In 2015 (Fig. 1a), the optimum N rate at stem elongation was 98 kg N ha⁻¹ at the conventional treatment (plus 40 kg N ha⁻¹ at tillering). In the organic treatments, the optimum N rate at stem elongation was approximately 118 kg N ha⁻¹. Maximum wheat grain yields were 8,456, 8,240 and 8,356 kg ha⁻¹ for conventional treatments, slurry and manure, respectively. Using organic amendments as initial fertilizers, approximately 20 kg N mineral ha⁻¹ less than at the conventional treatment was necessary for achieving the maximum wheat grain yield. The economically optimal dose at stem elongation was approximately the same (98 kg N ha⁻¹ for conventional treatment and 117 kg N ha⁻¹ for organics amendments).

In 2016, (Fig. 1b) the optimum N rate at stem elongation in the conventional treatment was 109 kg N ha⁻¹ (plus 40 kg N ha⁻¹ at tillering). In the organic treatments, the optimum N rate as slurry was 98 kg N ha⁻¹ and as manure was 147 kg N ha⁻¹. Maximum wheat grain yields were 10,227, 10,271 and 10,723 kg ha⁻¹ for conventional, slurry and manure treatments, respectively. When applying slurry as initial fertilizer, 51 kg N mineral ha⁻¹ less than the conventional treatment, were necessary for achieving the maximum wheat grain yield. Moreover, using manure as initial fertilizer, the same mineral N dose as at conventional treatment was necessary for achieving the maximum wheat grain yield, even though the maximum yield was approximately 500 kg ha⁻¹ higher. The economically optimal dose at stem elongation was approximately the same (110, 100 and 141 kg N ha⁻¹ for conventional, slurry and manure initial treatments, respectively).

In 2017, (Fig. 2c) the optimum N rate at stem elongation at the slurry treatment was 128 kg N ha⁻¹ and at manure was 156 kg N ha⁻¹. At conventional treatment, maximum yield was not achieved. Therefore, the maximum rate applied at stem elongation (160 kg N ha⁻¹) was taken as the optimum N rate at stem elongation. Maximum wheat grain yields were 5,841 and 6,205 kg ha⁻¹ for slurry and manure treatments, respectively. The economically optimal dose at stem elongation was lower than the optimum N rate at stem elongation (131, 111 and 130 kg N ha⁻¹ for conventional, slurry and manure initial treatments, respectively).

It is remarkable that in 2016 the previous crop was rapessed (*Brassica napus*). It has been shown that in the humid Mediterranean region of Spain, including rapeseed in the crop rotation increases wheat yields by about 10 % (ITGA 2009). Gallejones et al. (2012) concluded that rapeseed as the preceding crop for the wheat probably caused a higher N mineralization.

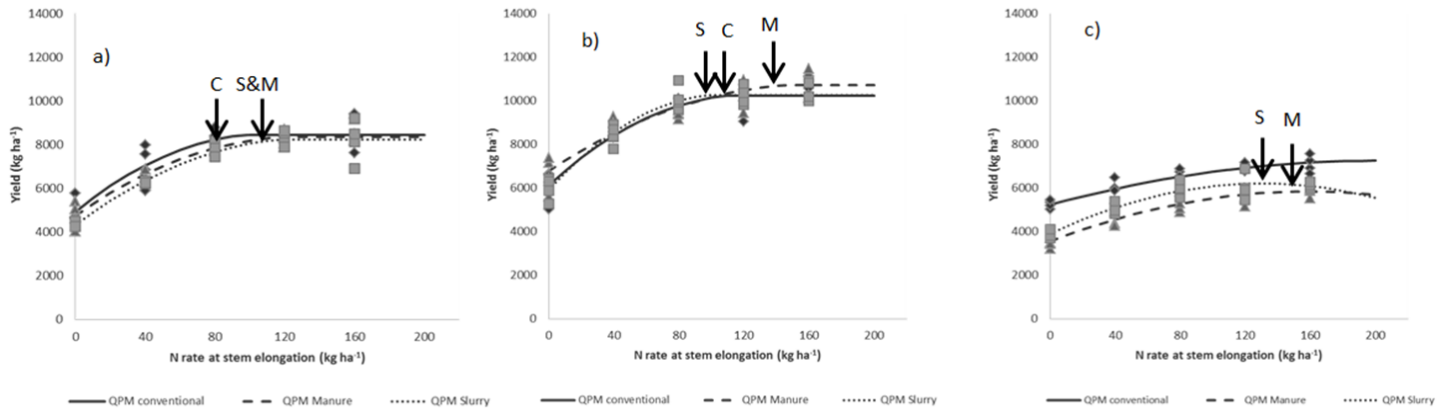


Fig. 1 Effect of N fertilization rate at stem elongation (GS-30) on yield (kg ha^{-1}) in 2015 (a), 2016 (b) and 2017 (c) wheat growing seasons respective to initial fertilization: conventional, dairy slurry, sheep manure. Quadratic plateau model was used for studying the yield response. C represents N fertilization rate when maximum yield was achieved by conventional treatment. S represents N fertilization rate when maximum yield was achieved by dairy slurry, and M represents N fertilization rate when maximum yield was achieved by sheep manure treatment

Soil mineral nitrogen (N_{\min})

Soil N supply is estimated through the assessment of soil mineral N content at the end of winter and it takes into account the net mineralization of soil organic matter, weather conditions and applied organic compounds. Soil mineral nitrogen depends on the net mineralization and crop uptake.

Before sowing, soil mineral nitrogen in 2015 was 50 kg N ha^{-1} , in 2016 was 42 kg N ha^{-1} and in 2017 was 34 kg N ha^{-1} (Table 2). At the end of winter (GS-21, beginning of tillering), N_{\min} values (Table 2) were lower in 2015 than in 2016 and 2017 in all initial fertilization treatments. The heavy rains between sowing and the end of winter (GS-21; Table 3) could explain N_{\min} low values during 2015 (Table 2). Arregui and Quemada (2006), in similar climate conditions (humid Mediterranean; Papadakis (1966)) showed that from sowing to mid-tillering (GS- 25) due to the high rainfall, low evapotranspiration and low crop demand, most of the mineral N present in the soil before sowing was lost by nitrate leaching. During the 2016 and 2017 wheat growing seasons the humid conditions (Table 3) allowed higher N_{\min} values (around 30 kg N ha^{-1}) at the end of winter (Table 2).

In 2015 at GS-21, conventional treatment had significantly higher N_{\min} values than dairy slurry and sheep manure (Table 2). The lower N_{\min} values at the treatments where organics were applied in 2015 could be explained due to immobilization (Mohanty et al. 2013). At the beginning of stem elongation (GS-30), N_{\min} values in 2016 were very low (Table 2). That could be explained by heavy rains (Table 3) and the high crop N need between the end of winter (GS-21) and stem elongation (GS-30).

Table 2. Soil Nmin content (kg N ha⁻¹; 0-30 cm) at the begging of the three growing seasons (2015, 2016 and 2017), beginning of tillering (GS-21) and beginnig of stem elonfation (GS-30) at Arkaute.

Growing season	Treatments	Nmin (0-30cm; kg N ha ⁻¹)		
		Initial	GS-21	GS-30
2015	ON			12 c
	40+ON	50	22 A b	13 b
	DS+ON		4 B b	13
	SM+ON		9 A b	12a
ON				1 c
2016	40+ON	42	30 a	3 b
	DS+ON		32 a	4
	SM+ON		30 a	1 b
	ON			
2017	40+ON	34	33 a	32 a
	DS+ON		36 a	16
	SM+ON		16 ab	14 a
	ON			

Means followed by a different capital letter indicate significant differences among initial treatments for each year (Duncan, $p > 0.05$); Means followed by a different small letter indicate differences among each initial treatment for different years

Soil characteristics also have an effect on N mineralization. Chantigny et al. (2004) suggested that clay fixation may have a significant effect on N availability to the crop. Other studies demonstrated that there is a significant interaction between manure and soil regarding the net mineralization and that the net N mineralization of cattle manures, cattle slurry and plant recovery is lower in clayey soils associated with the clay fixation of NH₄⁺-N (Shah et al. 2013). Mineralogical properties of the soil were analysed and it presented 40 % of phyllosilicate (Clays < 2 µm: Illite (%) = 30; Kaolinite (%) = 12; Smectite (%) = 58).

Table 3. Total rainfall (mm) and days elapsed between wheat growth stages (Zadoks, 1974) in three growing season (2015, 2016 and 2017) at Arkaute.

Growing season	Growth stage	Total rainfall (mm)	Days elapsed
2015	Sowing (24/11) - GS21 (09/03)	521	106
	GS21 (09/03) - GS30 (04/04)	95	30
	GS30 (04/04) - GS32 (29/04)	43	21
	GS32 (29/04) - GS37 (11/05)	6	12
2016	Sowing (06/11) - GS21 (19/01)	168	74
	GS21 (19/01) - GS30 (17/03)	296	56
	GS30 (17/03) - GS32 (30/03)	17	13
	GS32 (30/03) - GS37 (06/04)	24	7
2017	Sowing (18/11) - GS21 (02/03)	271	105
	GS21 (02/03) - GS30 (06/04)	58	35
	GS30 (06/04) - GS32 (12/04)	0	6
	GS32 (12/04) - GS37 (25/04)	0	13

The recommended rate of N fertilizer was calculated by the predicted N demand for the target yield minus the measured soil N_{\min} value. Target yields are highly variable and depend on spring weather. Although N_{\min} values at end of winter (GS-21) in 2016 and 2017 were similar, yields achieved in 2016 (10,200 – 10,700) were higher than yields in 2017 (5,800 – 6,200; Fig. 1). Therefore, the method has limitations and implies laborious and expensive sampling and analysis (Sylvester-Bradley et al. 2009). Ravier et al., 2016 highlighted many sources of uncertainty in soil N analysis that led to the exclusion of decision rules based on the monitoring of soil mineral content.

Proxy tools

The values of RapidScan CS-45 and N-tester™ were significantly higher in 2016 (Fig. 2b) than in 2015 and 2017 (Fig.2 a & c) in dairy slurry and sheep manure treatments. Having higher values in 2016 meant that the plant nitrogen nutritional status was better in 2016 than in 2015 and 2017. However, in conventional treatment values were similar among years (87 - 92 %). In 2015 (Fig.2a), the measurements (as a percentage compared to the overfertilized (280N) treatment) of both tools in all initial fertilization treatments were similar, showing differences between conventional treatment (88 %) and initial organic fertilization treatments (65 %). In 2016 (Fig. 2b) Yara N-tester™ showed differences between conventional treatment (92 %) and slurry treatment (85 %), but the tool didn't detect differences between manure (86 %) and the other two treatments. RapidScan CS-45 showed values between 84 - 88 %, but there were not differences among the three treatments. In 2017 (Fig. 3c), the measurements of both tools detected differences between conventional treatment (83 - 85 %) and initial organic fertilization treatments (60 - 65 %).

In the case of the dairy slurry treatment, when both proxy tool values were 60 - 65 % the optimum N rate at stem elongation for achieving the maximum yield was 118 - 128 kg N ha⁻¹. In dairy slurry and conventional treatments when readings were 85 - 90 % the optimum N rate was 100 - 110 kg N ha⁻¹. At the sheep manure treatment there was not a clear relationship between sensors values and optimum N rate. When values were 60 - 65 %, N recommendation was 117 or 155 kg N ha⁻¹ and 147 kg N ha⁻¹ for readings around 89%, probably caused by a lower ready available N (Defra 2017) and a different N mineralization pattern.

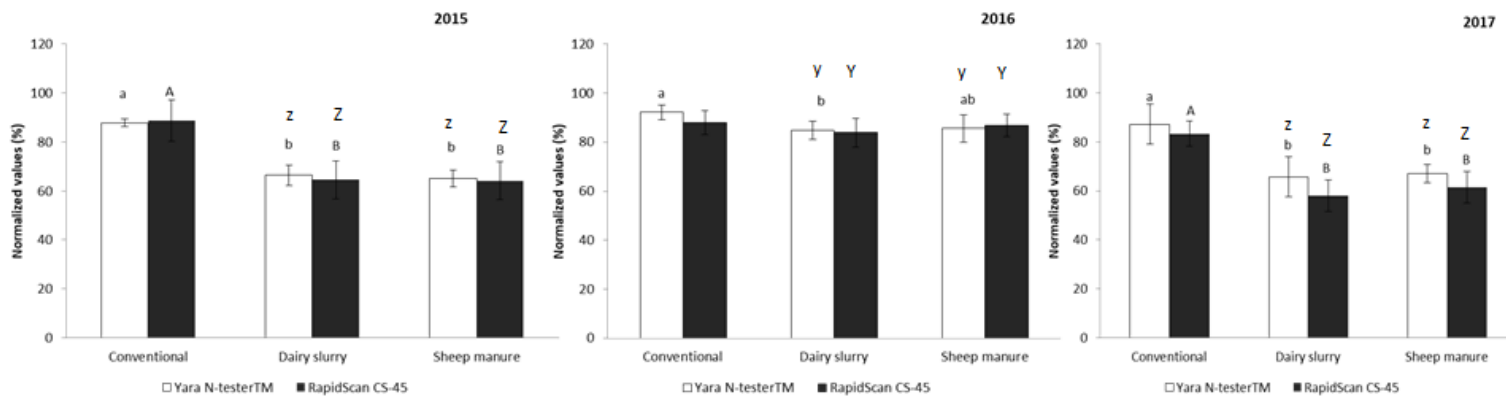


Fig. 2 Relation between initial fertilization (conventional, dairy slurry and sheep manure) and values obtained with tools for the diagnosis of N nutritional status (RapidScan CS-45, Yara N-tester™) at stem elongation (GS-30) in 2015 (a) 2016 (b) and 2017 (c) wheat growing seasons. Values were normalized assigning the 100% value to overfertilized (280 N) plot. Means with different small letters (a-b) represent significant differences among initial treatments in Yara N-tester™ measurements for each year, means with different capital letters (A-B) represent significant differences among initial treatments in RapidScan CS-45 measurements for each year (Duncan, $p > 0.05$), means with different small letters (y-z) represent significant differences among three years in Yara N-tester™ measurements for each treatment (Duncan, $p > 0.05$), and means with different capital letters (Y-Z) represent significant differences among the three years for RapidScan CS-45 measurements in each treatment (Duncan, $p > 0.05$)

These preliminary results look promising in order to adjust the N application rate at stem elongation with the Yara N-tester™ and RapidScan CS-45 readings when applying dairy slurry before sowing. However, at the sheep manure treatment, it is more difficult to find a relationship between sensor readings and yield. Both handheld N diagnostic tools were able to detect differences among nitrogen nutritional status in plants. This fact was supported by Arregui et al. (2006) and Ortuzar-Iragorri et al. (2005), who observed that chlorophyll meters enabled the prediction of the nitrogen status of wheat plants. Furthermore, Mullen et al. (2003) showed that in-season N demand for added N fertilizer in winter wheat could be detected using NDVI readings collected at stem elongation (GS-30). Moreover, algorithms using crop canopy reflectance sensing to make N recommendation for wheat have been identified, and it has been shown that active canopy sensors could be used in determining variable N rate applications in wheat from the mid-growing season (Raun et al. 2005; Reussi-Calvo et al. 2015). Marti et al. 2007 found significant correlations between NDVI, yield and biomass in wheat. On the other hand, Sylvester-Bradley et al. (2009) developed an alternative strategy for signaling soil nitrogen status and observed that NDVI of young canopies can signal soil N status where N_{min} is lower than 120-140 kg N ha⁻¹. In our case, both proxy tools showed a greater sensitivity than N_{min} when differentiating the initial fertilization treatments applied at the field trial. All these findings were very promising because the manner of fertilization could be changed with the aim of having a more precise N rate adjusted to the wheat crop demand. It should be noted that measurements taken with RapidScan CS-45 were less time-consuming than with Yara N-tester™ and were taken on the plant canopy and not just in the uppermost fully expanded leaf as with

chlorophyll meters. Thus, samples taken with RapidScan CS-45 can better represent the spatial variation of the crop N status. However, both tools should be calibrated for different genotypes or varieties, environmental conditions and growth stages to calculate the best possible N recommendation. Hence, plant sensing techniques to diagnose crop N status are usually based on the vegetation index used in conjunction with a reference area on the field with nonlimited N supply (Diacono et al. 2013). Ravier et al., 2017b suggested that is essential to ensure that the well-fertilized area is not N deficient, even after large amounts of nitrogen.

These hand-held tools are used in a mid-moment of the wheat growth period (GS-30), but many environmental variables (rainfall, temperature or relative humidity) affect the crop growth and development after this moment until harvest. However, any method of diagnosing N nutritional status at a particular moment has the same limitation. Since remote sensing measurements are not invasive and can be repeated several times during the growth period, information obtained on crop N status dynamics can be used for decision making in N fertilizer management. It has been shown that it is always possible to correct a nitrogen deficiency until the end of the cereal growth season (GS-65; Ravier et al. 2017a) if soil is wet (Soenen et al. 2017).

The application of N at the beginning of tillering (GS-21)

The 40 kg N ha⁻¹ application at the end of winter (GS-21) can be avoided when organic manures are applied according to results shown in 2015 and 2016, even when N_{min} at the end of winter was very low (4 – 9 kg N ha⁻¹) as in 2015 (Table 2). Thus, when the 40 kg N ha⁻¹ application at the beginning of tillering (GS-21) was not done in the treatments with organics as initial fertilizers, maximum yields were comparable in organics and conventional in 2015 and 2016 (Fig. 1a & b). However, in 2017 in the conventional treatment yields were higher than in organic treatments for each individual N rate at stem elongation (Fig.1c). It is important to repair that following mineral N application at stem elongation (GS-30) in 2017, it did not rain until flag leaf emergence (GS-37; Table 3). When there was not N application (0N; Table1) at stem elongation (GS-30), conventional treatment produced 1300 - 1700 kg ha⁻¹ more than dairy slurry and sheep manure treatments (Fig. 3c). Therefore, in that case, the 40 kg N ha⁻¹ application at tillering enhanced yield potential allowing higher productions. Basso et al. (2012) showed that wheat yield production in Mediterranean environment is highly affected by rainfall and amount of soil water stored into soil before and during the growing season. In the area where the study was carried out (humid Mediterranean (Papadakis (1966)) two of ten years have dry wheat growing season.

Conclusion

- Experimental findings to date have shown that Yara N-tester™ or Rapidscan CS-45 readings look promising in order to adjust the N application rate at stem elongation when applying dairy slurry before sowing. Thus, when both proxy tool values in dairy slurry were 60 - 65 % the optimum N rate at stem elongation for achieving the maximum yield was 118 - 128 kg N ha⁻¹. In dairy slurry and conventional treatments when readings were 85 - 90 % the optimum N rate was 100 - 110 kg N ha⁻¹. However, at the sheep manure treatment, it is more difficult to find a relationship between sensor readings and yield.
- When organic fertilizers were applied before sowing, the usual first N topdressing at the beginning of tillering could be avoided years with humid spring, suggesting new possibilities for a later application that might improve the protein content with lower likely fertilization costs. With a precise weather forecast it would be possible to time the N application with crop N demand and have a greater flexibility in decisions about when to apply N fertilizer.

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