# Late-Season Nitrogen Increases Improver Common and Durum Wheat Quality

Massimo Blandino,\* Patrizia Vaccino, and Amedeo Reyneri

#### **ABSTRACT**

The foliar N application at a low rate is a strategy that could be applied by farmers in substitution to the traditional late soil application when there are restrictions on N distribution. The aim of this study was to compare the effect of late-season N fertilization strategies on wheat (*Triticum* spp.) quality. Field experiments were set up in two different soils, in Northwest Italy. The effect of N fertilization was evaluated on two common (*T. aestivum* L.) and two durum wheat (*T. turgidum* L. var. *durum* Desf.) cultivars. Late-season N was soil-applied (40 kg N ha<sup>-1</sup>) as ammonium nitrate at heading, or foliar sprayed (5 kg N ha<sup>-1</sup>) at flowering, and compared with a control without N fertilization after the vegetative growth stage. Nitrogen soil application increased grain protein content (GPC) (+15%) for both crops. This fertilization strategy led also to higher test weight (+1.4%), kernel hardness (+14%), dough strength (W) (+42%), and lower tenacity/extensibility ratio P/L (-18%) in the common wheat, and to a lower yellowberry percentage in the durum wheat (-29%). The strategy to apply a foliar N fertilizer at anthesis at a lower rate, although it has a greater cost effectiveness, led to a less effective bread- or pasta-making quality enhancement, with an average increase of GPC and W only in the silt loam soil by 5 and 18%, respectively. This strategy could lead to more successful increase of GPC in soils with a probable lower nitrate leaching during the vegetative stages.

The wheat grain protein content is one of the key quality factors that can influence the end-use of wheat market classes throughout the world (Foca et al., 2007). High protein levels, which are linked with high kernel hardness, gluten strength and bread- and pasta-making quality, are desirable for improver common wheat (hard red wheat, HRW) and durum wheat (Brown and Petrie, 2006; Tronsmo et al., 2003). According to the established contracts, if the producers are able to satisfy and even surpass the GPC requirements, they enhance the possibility to sell the product to the supply chain, obtaining a premium price.

Nitrogen nutrition is widely considered as the main factor that affects storage proteins as well as the technological quality of grain (Wieser and Seilmeier, 1998). Low protein content can be attributed to N stress (Ayoub et al., 1994). However, since GPC can increase whenever N is available to the plant during grain filling, applying sufficient N to maximize the yield is not likely sufficient to avoid low protein discounts (Brown et al., 2005).

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Thus, once the N requirement for yield has been satisfied the distribution of additional N (30 – 50 kg N  $ha^{-1}$ ), after the vegetative growth stage (GS, Zadoks et al., 1974), is a common practice to increase wheat protein in irrigated cropping systems or in climatic zones with sufficient spring rainfall (Borghi et al., 1995; Brown and Petrie, 2006; López-Bellido et al., 1998). Fuertes-Mendizábal et al. (2010) reported that not only increasing the N fertilization rate, but also splitting the N fertilization into two or more soil amendments had a beneficial effect on GPC and on wheat rheological quality. Timing the N application to correspond to reproductive development leads to a higher protein synthesis and storage in grains, but delaying fertilization too long may restrict the amount of N that can be converted into quality protein (Brown et al., 2005). Nitrogen applied between boot and anthesis could be effective in enhancing GPC, although they do not generally increase grain yield (Borghi et al., 1995). Taking into account different application timings, Brown et al. (2005) and Ayoub et al. (1994) reported that N soil fertilization at heading resulted in a higher flour protein concentration; moreover, Ottman et al. (2000) and Jones and Olson-Rutz (2012) indicated that this late-season soil application should be in the 30 to 50 kg N ha<sup>-1</sup> range. Ammonium nitrate may be a better source for the top-dressed granular application of N than urea, as it is less subject to volatilization losses (Bremner, 1995). The risk of foliage burning using granular application of ammonium nitrate is low.

Abbreviations: AULGC, area under the leaf greenness curve; CEC, cation exchange capacity; FHB, Fusarium head blight; GDDs, growing degree days; GPC, grain protein content; GS, growth stage; HNT, hydro N-tester; NVZ, nitrate vulnerable zones; P/L, tenacity/extensibility ratio; TW, test weight; TKW, thousand kernel weight; YB, yellowberry; W, dough strength.

Another strategy that can be adopted to enhance wheat quality, in terms of grain protein, is to use concentrated foliar sprays between heading and flowering (Fageria et al., 2009), and these are often applied in mixtures with fungicides to control Fusarium head blight (FHB).

Gooding and Davies (1992) suggested that benefits include reduced denitrification and leaching, improved fertilizer efficiency and reinforce the late seasonal N uptake. Tea et al. (2004) demonstrated that N provided by foliar fertilization is mainly incorporated in storage proteins, which are responsible for bread-making quality. Moreover, this late-season N fertilization strategy could be more easily adapted by farmers as it does not require a dedicated passage in the field, if combined and tank mixed with a fungicide treatment, although there is a limit to how much foliar N can be applied without incurring leaf burn (Jones and Olson-Rutz, 2012; Krogmeier et al., 1989). The risk of foliage burning is higher when the N source is ammonium nitrate or ammonium sulfate rather than urea (Gooding and Davies, 1992). Moreover, foliar application needs to be avoided early in the morning when dew is still on the crop or when there are high temperatures during the day (Arnall et al., 2012; Woolfolk et al., 2002). Leaf burn can be severe: in this case, the protein enhancement after N foliar application could occur as a result of yield suppression due to leaf burning (Brown and Petrie, 2006).

The optimum timing of N spraying on wheat has shown that post-pollination foliar N leads to the largest grain protein improvement (Bly and Woodard, 2003; Tea et al., 2007; Woolfolk et al., 2002). A positive relationship between N, provided by foliar fertilization at anthesis, and the bread-making quality of flour has been observed (Blandino and Reyneri, 2009; Tea et al., 2004). In greenhouse experiments, the recovery of <sup>15</sup>N-labeled N in wheat grain is generally higher for soil applications than foliar ones, although conflicting results have been observed on their capacity to increase the final GPC (Alkier et al., 1972; Rawluk et al., 2000). However, Gooding et al. (2007) reported a higher recovery of N in grains in field experiments, after urea foliar application, compared to soil fertilization with ammonium nitrate.

Even though the qualitative effect of late-season fertilizer application on wheat quality has been studied extensively, it has not been directly established in the field if a foliar application of N at a lower rate could produce a comparable protein increase than soil placement strategy at higher N rate, which required a dedicated passage and which is not suitable in nitrate vulnerable zones (NVZ, according to the European Union nitrates directive 91/676/EEC, EEC, 1991) or when there are other restrictions on the rate of N application. Moreover, the GPC increase for late-season N is quite variable and several factors, other than N management, can influence the protein response in field conditions, such as yield potential (Oury and Godin, 2007) and growing conditions from heading to the end of grain filling, with an important role played by the N and water availability, the occurrence of disease attack, and other stresses (Dimmock and Gooding, 2002).

The aim of this study was to compare the effect of late-season N fertilization strategies through granular top-dressed soil or foliar applications on different productive and quality parameters of improver common wheat and durum wheat, with

different soils and growing conditions. No complete comparison of the fertilization strategies actually applied by farmers to manage late-season N nutrition in different cropping systems have been previously reported, considering their impact also on flour quality. The objective was to develop rational N management practices for temperate areas to achieve the quality required for these market categories by supply chains.

#### **MATERIALS AND METHODS**

# **Experimental Site and Treatments**

The study was performed in Northwest Italian plain (Piedmont Region), during two growing seasons (2008–2009 and 2009–2010). The experiment was performed at two soils, sandy loam and silt loam (Table 1). Soil was sampled from 0 to 60 cm using Eijkelkamp cylindrical augers at wheat tillering (GS 23). The sandy loam and the silt loam soil are characterized by a medium-low and medium-high cation exchange capacity (CEC), and therefore by a probable high and low N leaching during the growing season, respectively.

Three different late-season N fertilization strategies have been compared:

- T1, control without N application after the stem elongation stage;
- T2, granular N soil fertilization at the beginning of heading (GS 52): 40 kg N ha<sup>-1</sup> was applied top-dressed as ammonium nitrate (27% N w/w);
- T3, foliar liquid N fertilization at anthesis (GS 65):
   5 kg N ha<sup>-1</sup> was applied using the foliar liquid fertilizer YaraVita Last N (Yara Italia S.p.A., Milano, Italy; composition 312 g N L<sup>-1</sup>, 25%, of which 3.9, 3.9, and 10.6% as nitrate, ammonium, and ureic N, respectively; 16 L of product formulation ha<sup>-1</sup>, according to the commercial recommended rate).

The comparison of late-season fertilizer followed the technical solution most frequently applied by wheat growers in Europe. In particular the foliar application at a low N rate is the late-season fertilization commonly applied in NVZ or when other restrictions on N fertilization take place. Moreover, the foliar liquid N fertilization could be easily and more economically adapted by farmers if combined with the fungicide treatment at anthesis.

Both granular top-dressed soil or foliar late-season N fertilization were applied midday, to be practical also on a large scale.

A total of  $130 \text{ kg N ha}^{-1}$  was applied, from tillering to the beginning of the stem elongation stage, to all the compared treatments (T1, T2, T3) as a granular ammonium nitrate fertilizer, split  $50 \text{ kg N ha}^{-1}$  at GS 23 and  $80 \text{ kg N ha}^{-1}$  at GS 32. The sowing, harvesting, and N fertilization dates are reported in Table 2, for each year and each site.

An untreated control that did not receive any mineral N fertilizer during the growing season was introduced as a control of soil fertility, reporting the canopy greenness evolution during the grain-filling stage of the different growing seasons.

The compared treatments were applied to two common wheat and two durum wheat cultivars. The common wheat cultivars were Bologna (S.I.S. Società Italiana Sementi, San Lazzaro di Savena, BO, Italy) and Generale (Consorzio nazionale sementi, Conselice, Ra, Italy), which are classified, according to the Italian bread-making quality grade (Foca et al., 2007) as

Table I. Main physical and chemical characteristics of the soil for the field experiments carried out in the 2008 to 2010 period in Northwest Italy.

Parameters†	Measurement	Sandy Ioam	Silt loam
Site		Cigliano (VC)	Poirino (TO)
Geographical coordinates		45°19′ N, 8°01′ E	44°54′ N, 7°24′ E
Altitude	m	237	262
Soil (USDA classification)		Typic Hapludalf	Typic Udifluvent
and (2–0.05 mm)	%	50.7	20.5
Silt (0.05–0.002 mm)	%	38.9	64.7
Clay (<0.002 mm)	%	10.4	14.9
Bulk density	g cm <sup>-3</sup>	1.53	1.28
ЬН		6.2	6.9
Organic matter	%	1.47	1.25
Organic C	%	0.85	0.73
Total N	%	0.079	0.066
C/N		10.7	11.0
Cation exchange capacity (CEC)	cmol <sub>c</sub> kg <sup>-1</sup>	0.92	1.45
Exchangeable K	mg kg <sup>-1</sup>	76	78
Available P	mg kg <sup>-l</sup>	28	20
Total N			
2008–2009 growing season	%	0.077	0.062
2009–2010 growing season	%	0.080	0.070

<sup>†</sup> With the exception of Total N, the reported parameter values are the average data of two growing seasons (2008–2009 and 2009–2010).

improver and superior bread making-quality wheat, respectively. The durum wheat varieties were Dylan (Apsovsementi S.p.A., Voghera, PV, Italy) and Saragolla (Produttori Sementi Bologna, Argelato, BO, Italy), which are classified as high and medium quality wheat, respectively.

The treatments were assigned to experimental units using a completely randomized block design with four replicates. The plot size was 7 by 1.5 m. The plots were seeded after an autumn plowing (30 cm) and disk harrowing to prepare a suitable seedbed, following a previous maize (*Zea mays* L.) crop for grain. Planting was conducted in 12 cm wide rows in October or November at a seeding rate of 450 seeds m $^{-2}$ . The experimental field received 115 kg ha $^{-1}$  of  $K_2O$  each year. The weed control was conducted with isoproturon (3-(4-isopropylphenyl)-1,1-dimethylurea or 3-p-cumenyl-1,1-dimethylurea) and diflufenican (2',4'-difluoro-2-( $\alpha,\alpha,\alpha$ -trifluoro-m-tolyloxy)nicotinanilide) at wheat tillering (GS 23).

All the plots were treated with two applications of fungicide: a mixture of azoxystrobin (methyl (E)-2-{2 [6-(2-cyanophenoxy) pyrimidin-4-yloxy]phenyl}-3-methoxyacrylate) and cyproconazole ((2RS,3RS;2SR,3SR)-2-(4-chlorophenyl)-3-cyclopropyl-1-(1H-1,2,4-triazol-1-yl)butan-2-ol) (Amistar Xtra, Syngenta Crop Protection S.p.A., Milan, Italy) applied at 0.2 kg + 0.08 kg a.i. ha<sup>-1</sup> at steam elongation (GS 34) to control foliar

disease, and a mixture of cyproconazole and procloraz (N-Propyl-N-[2-(2,4,6-trichlorophenoxy)ethyl]-1H-imidazole-1-carboxamide) (Tiptor Xcell, Syngenta Crop Protection S.p.A., Milan, Italy) applied at 0.02~kg+0.17~kg a.i. ha $^{-1}$  at heading (GS 55) to avoid Fusarium Head Blight infection and protect flag leaf greenness. The fungicides, or the leaf N fertilizer, were applied using a three nozzle precision sprayer (T-Jet 110/04) emitting a fine mist at a slow walk to ensure effective coverage. The delivery pressure at the nozzle head was 324 KPa.

The grain yields were obtained by harvesting with a Walter Wintersteiger cereal plot combine-harvester. The grain yield results were adjusted to a 13% moisture content and 2 kg grain samples were taken from each plot for the qualitative analyses.

### Flag Leaf Greenness

A chlorophyll meter, hydro N-tester (HNT) (Hydro-Agri, now Yara) was used to measure the relative flag leaf greenness after the late N fertilization.

The HNT is a hand-held instrument that measures the light transmitted by a plant leaf at two different wavelengths (650 and 960 nm) (Arregui et al., 2006). The ratio of the light transmitted at these wavelengths, in addition to the ratio determined with no sample, is processed by the instrument to produce a digital reading. The HNT values are numerical,

Table. 2. Main trial information and date of N fertilization in the field experiments carried out in the 2008 to 2010 period in Northwest Italy.

Trial information	Growing stage		Dat	ie .	
Growing seasons		2008-	-2009	2009	2-2010
Soil		sandy loam	silt loam	sandy loam	silt loam
Sowing date		17 Nov. 2008	10 Nov. 2008	4 Nov. 2009	30 Oct. 2009
N fertilization	tillering (GS 23)	11 Mar. 2009	18 Mar. 2009	16 Mar. 2010	19 Mar. 2010
	stem elongation (GS 32)	8 Apr. 2009	17 Apr. 2009	14 Apr. 2010	20 Apr. 2010
	heading (GS 52)†	7 May 2009	8 May 2009	3 May 2010	10 May 2010
	foliar N application (GS 65)‡	18 May 2009	18 May 2009	17 May 2010	19 May 2010
Harvest date		30 June 2009	9 July 2009	8 July 2010	15 July 2010

<sup>†</sup> For only T2 treatment.

<sup>‡</sup> For only T3 treatment.

dimensionless values that are proportional to the amount of total chlorophyll present in the leaf (Peltonen et al., 1995).

Readings were taken using the HNT at mid-length of the flag leaf from 30 randomly selected plants per plot. The HNT measurements were performed at different growth stages: the beginning of heading, before the granular or foliar N fertilization (GS 52), the milk stage (GS 75) and the dough stage (GS 85).

The area under leaf greenness curve (AULGC) was calculated for each treatment starting from the HNT measurements conducted during crop maturation using the following formula:

AULGC = 
$$\sum_{i=1}^{n-1} \{ [(R_i + R_{i+1})/2](t_{i+1} - t_i) \}$$
 [1]

where  $R_i$  is the average HNT reading value of the ith record,  $R_{i+1}$  is the average HNT reading value of the i+ith record,  $t_{i+1} - t_i$  is the time of day between the ith record and the i+ith record and n is the number of observations.

# **Small-Scale Quality Analyses**

The test weight (TW) was determined by means of a Dickey-John GAC2000 grain analysis meter (Dickey-John Corp., Auburn, IL) according to the supplied program and after a validation with reference materials. The thousand kernel weight (TKW) was determined on two 100-kernel sets for each sample using an electronic balance.

Grain samples (50 g) from each plot were milled using a Retsch ZM 200 (Retsch GmbH, Haan, Germany), fitted with a 1 mm aperture sieve, and the resulting wholemeal was analyzed by near-infrared reflectance spectroscopy, using a NIRSystems 6500 monochromator (Foss-NIRSystems, Silver Spring, MD).

The grain protein content (GPC;  $N \times 5.7$ , dry matter basis) was determined for both the durum and common wheat according to AACC 39-10 (AACC, 2000); grain hardness was determined for common wheat only, according to AACC 39-70A. All the analyses were performed in duplicate.

The gluten index and the yellowberry incidence (YB) were recorded for the durum wheat samples. The gluten index quality was evaluated according to ICC-158 (ICC, 1992) using the Glutomatic System (Perten Instruments, Hägersten, Sweden). The YB was determined by counting the percentage of mealy kernels after a visual inspection of four slabs with 40 grains, cut thought the granotomo of Pohl.

#### Large-Scale Quality Analyses

Only cultivar Bologna was taken into consideration for the large-scale analyses of the common wheat, since it is the reference cultivar for Italian milling due to its huge diffusion on the Italian market. The four replicates for each entry unit (treatment) were bulked and milled with an experimental Bona 4RB mill (Bona, Monza, Italy) after tempering according to their hardness. The rheological properties of the flours were evaluated using a Chopin alveograph, according to ICC-121, and a Brabender farinograph, according to ICC-115-D (ICC, 1992).

## **Statistical Analysis**

The normal distribution and homogeneity of variances were verified for each trial by performing the Kolmogorov–Smirnov

normality test and the Levene test, respectively. All the recorded parameters were compared by means of an ANOVA, in which the treatment and the cultivars were the independent variables. The ANOVA was conducted separately for each year and each site combination and for the common and durum wheat, to satisfy the homogeneity of variance assumption. Multiple comparison tests were performed according to the Ryan–Einot–Gabriel–Welsh F (REGWF) test on treatment means (Bender and Langeb, 2001). The statistical package SPSS for Windows, Version 19.0 (SPSS Inc., Chicago) was used for the statistical analysis.

### **RESULTS**

# **Weather Conditions**

The two growing seasons differed mainly as far as the total rainfall that occurred in the period between the wheat sowing (November) and the booting stage (April) is concerned (Table 3). During this period, the 2008–2009 growing season experienced the greatest total rainfall (>750 mm in both sites), while the total rainfall in 2009–2010 during the wheat vegetative stages was lower than 450 mm. On the other hand, the 2009–2010 growing season reported higher rainfall than the 2008–2009 period from wheat heading to the end of the ripening stages (from May to June). The total GDD was higher in the 2008–2009 period than in 2009–2010.

The total rainfall and GDD also differed from site to site, with the highest level of rainfall and GDD consistently observed in the sandy loam site (Cigliano) for both growing seasons. In particular, the GDD in May and June, from the flowering to the end of ripening, was always higher in Cigliano site than in Poirino.

#### **Leaf Greenness**

No leaf burn or other damages to wheat were recorded for the granular top-dressed soil or foliar application. Both lateseason fertilization strategies were applied using the tram lines previously established.

The response of the common and durum wheat to the applied N fertilization strategies were different for the different sites (Tables 4 and 5). At the two sites and in both growing season, and for both the common and durum wheat, the granular fertilization of 40 kg N ha<sup>-1</sup> at the beginning of heading (T2) increased the AULGC value when compared to the control (T1). The N foliar application at flowering (T3) to the common wheat significantly increased AULGC in both growing seasons at the silt loam soil but only in the 2009–2010 period at the sandy loam soil site. The AULGC values of durum wheat were higher in the T3 treatment than the T1 in the 2009–2010 growing season at the sandy loam soil site and at the silt loam soil site in 2008–2009.

The granular N fertilization at the beginning of heading (T2), showed higher AULGC values in each experiment, compared to the foliar N fertilization at anthesis (T3), with the exception of the trial set up for common wheat in the sandy loam soil in both growing seasons.

The AULGC of the compared common wheat cultivars was only significantly different in the experiment conducted in the 2008–2009 growing season at the silt loam soil site (cultivar Bologna > cultivar Generale). Durum wheat Saragolla only

Table. 3. Monthly rainfall and growing degree days (GDD 0s) from sowing (November) to the end of ripening (June) in the 2008 to 2010 period in the studied sites.†

			2008–2009		2009–2010				
Soil	Month	Rainfall	Rainy days	GDD‡	Rainfall	Rainy days	GDD†		
		mm	no.	$\Sigma$ °C d $^{-1}$	mm	no.	$\Sigma$ °C d <sup>-1</sup>		
Sandy loam (Cigliano)	November	199	13	238	103	6	236		
	December	250	11	126	53	7	105		
	January	46	6	88	47	7	58		
	February	74	6	155	96	14	115		
	March	113	8	292	67	9	245		
	April	253	13	420	54	9	398		
	May	91	4	599	185	12	510		
	June	108	8	651	118	9	638		
	November- June	1134	69	2567	724	73	2305		
	November-April	935	57	1317	420	52	1157		
	May-June	199	12	1250	303	21	1148		
Silt loam (Poirino)	November	160	10	225	79	6	223		
	December	188	11	112	35	8	92		
	January	50	9	62	56	8	39		
	February	37	6	151	79	12	96		
	March	85	7	286	58	9	228		
	April	242	15	379	45	7	381		
	May	26	4	584	108	11	487		
	June	95	5	635	140	9	604		
	November-June	882	67	2433	600	70	2149		
	November-April	761	58	1214	352	50	1058		
	May-June	121	9	1219	248	20	1091		

<sup>†</sup> Source: Rete Agrometeorologica del Piemonte-Regione Piemonte-Assessorato Agricoltura- Settore Fitosanitario, sezione di Agrometeorologia.

showed significantly higher leaf greenness during the maturation stages in the 2009–2010 growing season at the sandy loam soil site. The interactions between late-season N fertilization and the cultivars were never significant, for either the common or the durum wheat.

#### **Grain Yield and Yield Parameters**

Late season N did not influence yield or TKW in common wheat (Table 4). The TW of the wheat fertilized with a N foliar application at anthesis (T3) was only higher than T1 in the 2009–2010 experiment at the silt loam soil site.

The sandy loam soil resulted in a higher grain yield for cultivar Generale, while cultivar Bologna produced more in the silt loam soil. The TKW of cultivar Generale was always significantly higher than those of cultivar Bologna. With the exception of the trial performed in 2008–2009 at the sandy loam soil site, cultivar Bologna showed significantly higher TW than cultivar Generale. The interactions between late-season N fertilization and the common wheat cultivar was never significant for grain yield, TKW, or TW.

As far as the durum wheat is concerned, ANOVA only showed a significant effect of the late-season N fertilization on grain yield in the experiment conducted in the 2008–2009 growing season at the sandy loam soil site (Table 5): in this case granular N soil fertilization at heading increased grain yield, compared to foliar N application, by 10% (P < 0.05). Moreover, ANOVA did not show a significant effect of late-season N fertilization on TKW or TW (Table 5). With the exception of the 2009–2010 growing season at the silt loam soil site, no significant difference in grain yield between the two cultivars was observed, while cultivar Dylan showed significantly higher

TW than cultivar Saragolla at each site. The cultivar effect on TKW varied with the year and site. The interactions between late-season N fertilization and the durum wheat cultivar was never significant for grain yield, TKW, or TW.

# Grain Protein Content and Other Qualitative Parameters

The ANOVA showed a significant effect of late-season N fertilization on the GPC of the common (Table 4) and durum wheat (Table 5) in each experiment, but the response to the N fertilization strategies applied was different for the growing seasons and soils.

In all the experiments, the application of granular N soil fertilizer at heading (T2) increased GPC significantly (P < 0.001), compared to the control without late-season N fertilization (T1). On average, the GPC increased by 1.92 and 1.31% for common and durum wheat, respectively. On the other hand, the foliar N application (T3) on both crops, only significantly increased GPC, compared to the control without late-season N fertilization (T1), in the trials conducted in the silt loam soil, while the difference between T1 and T3 was never significant in the sandy loam soil.

The foliar N fertilizer in the silt loam soil, increased the GPC by 0.60 and 0.58% for common and durum wheat, respectively. Furthermore, the T2 treatment resulted in a significantly higher GPC than T3 in all the experiments.

The ANOVA showed a significant effect of cultivar on GPC in the sandy loam soil in both crops and for both growing seasons and at the silt loam soil site but only for durum wheat in 2008–2009 growing season. As expected, in these cases, cultivars Bologna and Dylan showed higher GPC than cultivars

<sup>‡</sup> GDD: Accumulated growing degree days for each month using a 0°C base.

Table. 4. Effect of late-season N applications on the area under leaf greenness curve (AULGC), grain yield, thousand kernel weight (TKW), test weight (TW), grain protein content (GPC), and hardness; field experiments were carried out on two common wheat cultivars in Northwest Italy in the 2008 to 2010 period.

Soil	Growing season	Factor	Source of variation	AULGC	Grain yield	TKW	TW	GPC	Hardness
				HNT†	t ha <sup>-l</sup>	g	t ha <sup>-l</sup>	%	
Sandy Ioam (Cigliano)	2008–2009	Late season	TI	15,768b	3.6a	35.8a	78.5b	12.3b	64b
		N (A)‡	T2	18,136a	3.7a	35.4a	80.0a	14.6a	71a
			T3	17,057ab	3.6a	35.8a	79.4ab	12.3b	65b
			P(F)	**	ns§	ns	*	***	**
			SEM¶	2,027	0.4	2.2	1.5	0.5	5.1
		Cultivar (B)#	Bologna	16,564a	3.3b	32.9b	79.5a	13.4a	71a
			Generale	17,410a	3.9a	38.5a	79.1a	12.7b	62b
			P(F)	ns	***	***	ns	***	***
			SEM¶	1,655	0.3	1.8	1.2	0.4	4.2
		A×B	P(F)	ns	ns	ns	ns	ns	ns
Sandy Ioam (Cigliano)	2009–2010	Late season	TI	21,015b	7.7a	38.8a	83.Ib	12.4 b	68b
		N (A)	T2	22,758a	7.6a	38.8a	84.0a	14.3 a	78a
			T3	22,167a	7.6a	39.8a	83.6ab	12.4 b	70b
			P(F)	***	ns	ns	**	***	***
			SEM	1,083	0.6	2.4	0.7	0.4	5.3
		Cultivar (B)	Bologna	21,833a	7.4b	36.2b	84.5a	13.6a	81a
			Generale	22,128a	7.9a	42.0a	82.6b	12.5b	63b
			P(F)	ns	**	***	***	***	***
			SEM	884	0.5	1.9	0.5	0.3	4.3
		A×B	P(F)	ns	ns	ns	ns	ns	ns
Silt Ioam (Poirino)	2008–2009	Late season	TI	14,673c	4.5a	35.5a	76.8b	11.5c	45c
		N (A)	T2	17,397a	4.7a	36.5a	77.9a	13.6a	55a
			T3	15,810b	4.4a	35.8a	77.1b	12.0b	49b
			P(F)	***	ns	ns	***	***	***
			SEM	1,246	0.4	2.2	0.6	0.4	5.1
		Cultivar (B)	Bologna	16,718a	4.7a	33.7b	78.2a	12.3a	54a
			Generale	15,201b	4.3b	38.2a	76.4b	12.4a	45b
			P(F)	***	**	***	***	ns	***
			SEM	1,017	0.3	1.8	0.5	0.3	4.2
		A×B	P(F)	ns	ns	ns	ns	ns	ns
Silt Ioam (Poirino)	2009–2010	Late season	TI	16,192b	4.8a	36.6a	79.9 b	13.3c	65b
		N (A)	T2	18,119a	5.0a	37.4a	80.8a	14.8a	72a
			T3	16,457b	4.9a	36.8a	80.7a	14.0b	68b
			P(F)	***	ns	ns	*	***	**
			SEM	1,332	0.6	2.1	0.9	0.6	5.6
		Cultivar (B)	Bologna	17,183a	5.la	33.4b	80.8a	14.0a	76a
			Generale	16,663a	4.6b	40.5a	80.1b	14.0a	61b
			P(F)	ns	**	***	*	ns	***
			SEM	1,088	0.5	1.7	0.7	0.5	4.6
		$A \times B$	P(F)	ns	ns	ns	ns	ns	ns

<sup>\*</sup> Means followed by different letters are significantly different at the 0.05 probability level.

<sup>\*\*</sup> Means followed by different letters are significantly different at the 0.01 probability level.

<sup>\*\*\*</sup> Means followed by different letters are significantly different at the 0.001 probability level.

<sup>†</sup> HNT, hydro N-tester.

<sup>‡</sup> The reported values of the late-season N application factor are based on 8 replications (two cultivars by four repetitions).

<sup>§</sup> ns: not significant at the 0.05 probability level.

<sup>¶</sup> SEM: standard error of mean.

<sup>#</sup> The reported values of the cultivar factor are based on 12 replications (three treatments by four repetitions).

Generale and Saragolla, respectively. The interactions between the late-season N fertilization and the cultivars were never significant, for either the common or the durum wheat.

Taking into account the level of 13.5% for GPC proposed in the Italian Synthetic Quality Index for common wheat classification with granular N soil fertilization at heading (T2), this quality requirement was satisfied in 94 and 88% of the cases in the sandy loam and silt loam soil, respectively. Instead the

specific grain protein goal was never achieved in the sandy loam soil with the N foliar fertilizer (T3) and only in 50% of the cases in the silt loam soil. Taking into account only the T1 and T3 treatments, a significant relationship (P < 0.05) between flag leaf greenness, expressed as AULGC values, and GPC has been observed in the silt loam soil ( $R^2 = 0.53$  and  $R^2 = 0.48$  for common and durum wheat, respectively), while the regression was never significant in the sandy loam soil.

Table. 5. Effect of late-season N applications on the area under leaf greenness curve (AULGC), grain yield, thousand kernel weight (TKW), test weight (TW), grain protein content (GPC), gluten index, and yellowberry percentage (YB); field experiments were carried out on two durum wheat cultivars in Northwest Italy in the 2008 to 2010 period.

Soil	Growing season	Factor	Source of variation	AULGC	Grain yield	TKW	TW	GPC	Gluten index	YB
				HNT†	t ha <sup>-1</sup>	g	t ha <sup>-I</sup>	%		%
Sandy Ioam (Cigliano)	2008–2009	Late season	TI	19,876b	4.4ab	50.3a	71.9a	12.2b	75.8a	35.3a
		N (A)‡	T2	22,582a	4.7a	51.la	71.7a	13. <del>4</del> a	72.8a	23.0b
			T3	20,488b	4.2b	50.0a	71.8a	12.4b	77.7a	27.8ab
			P(F)	***	*	ns§	ns	***	ns	***
			SEM¶	1.159	0.4	4.8	2.0	0.3	5.0	4.0
		Cultivar (B) #	Bologna	20,734a	4.5a	51.0a	72.7a	13.1a	57.0b	24.6b
			Generale	21,231a	4.4a	50.0a	70.8b	12.2b	93.9a	32.9a
			P(F)	ns	ns	ns	**	***	***	***
			SEM§	946	0.4	3.9	1.7	0.3	<b>4</b> . I	3.3
		$A \times B$	P(F)	ns	ns	ns	ns	ns	ns	ns
	2009–2010	Late season	TI	17,706c	6.3a	49.5a	76.3a	12.5b	85.9a	10.3a
		N (A)	T2	19,051a	6.3a	49.0a	76.5a	14.1a	88.5a	3.1b
			T3	18,331b	6.3a	49.3a	76.3a	12.8b	89.4a	5.9ab
			P(F)	*okok	ns	ns	ns	***	ns	***
			SEM	866	0.6	2.4	0.8	0.6	3.8	3.3
		Cultivar (B)	Bologna	18,061b	6.5a	48.1b	77.5a	13.4a	80.3b	5.3b
			Generale	18,664a	6.2a	50.4a	75.3b	12.8b	95.6a	7.6a
			P(F)	*	ns	*	***	***	slokek	*
			SEM	707	0.5	2.0	0.7	0.5	3.1	2.7
		A×B	P(F)	ns	ns	ns	ns	ns	ns	ns
Silt Ioam (Poirino)	2008–2009	Late season	ΤI	21,942c	2.9a	50.2a	73.3a	12.8c	78.0a	32.0a
		N (A)	T2	24,059a	3.2a	51.6a	73.7a	13.9a	81.2a	27.0a
			T3	23,078b	3.la	50.5a	73.4a	13.3b	81.2a	29.3a
			P(F)	***	ns	ns	ns	***	ns	ns
			SEM	1,375	0.5	1.9	0.6	0.3	5.2	5.0
		Cultivar (B)	Bologna	22,945a	3.0 a	52. <del>4</del> a	75.0a	13.9a	63.8b	25.0b
			Generale	23,107a	3.1 a	49.2b	72.0b	12.7b	96.4a	33.9a
			P(F)	ns	ns	***	***	***	***	***
			SEM	1,123	0.4	1.5	0.5	0.3	4.2	4.1
		$A \times B$	P(F)	ns	ns	ns	ns	ns	ns	ns
	2009–2010	Late season	TI	18,131b	3.8a	44.7a	70.7a	12.1c	94.3a	14.6a
		N (A)	T2	20,103a	3.9a	44.8a	71.0a	13.5a	91.6a	15.3a
			T3	18,763b	3.8a	45.2a	70.8a	12.8b	91.4a	17.5a
			P(F)	***	ns	ns	ns	***	ns	ns
			SEM	1324	0.5	1.5	1.0	0.6	4.2	6.6
		Cultivar (B)	Bologna	18,798a	4.3a	46.0a	71.5a	12.9a	90.3b	14.8a
		` '	Generale	19,200a	3.4b	43.8b	70.2b	12.6a	94.6a	16.8a
			P(F)	ns	***	***	***	ns	***	ns
			SEM	1,081	0.4	1.2	0.8	0.5	3.4	5.4
		A×B	P(F)	ns	ns	ns	ns	ns	ns	ns

<sup>\*</sup> Means followed by different letters are significantly different at the 0.05 probability level.

<sup>\*\*</sup> Means followed by different letters are significantly different at the 0.01 probability level.

<sup>\*\*\*</sup> Means followed by different letters are significantly different at the 0.001 probability level.

<sup>†</sup> HNT: hydro N-tester.

<sup>‡</sup> The reported values of the late-season N application factor are based on 8 replications (two cultivars by four repetitions).

<sup>§</sup> ns: not significant at the 0.05 probability level.

<sup>¶</sup> SEM: standard error of mean.

<sup>#</sup> The reported values of the cultivar factor are based on 12 replications (three treatments by four repetitions).

The ANOVA showed a significant effect of late-season N fertilization on common wheat grain hardness in each experiment (P < 0.01, Table 4). In the trials conducted in the sandy loam soil, only the granular N fertilization at heading significantly increased hardness compared to T1. In the trials conducted in the silt loam soil, grain hardness was significantly increased by the foliar N application (T3) compared to T1 (+7%) in the 2008–2009 growing season, but granular N fertilization (T2) led to a further significant increase (+8%). The interactions between late-season N fertilization and the cultivars were never significant.

The effect of late-season N fertilization on the rheological properties of the dough from cultivar Bologna is reported in Table 6. On average, and compared to the control (T1), the granular N fertilization at heading (T2) increased W by 36 and 47% in the experiments conducted at the sandy loam and silt loam soil sites, respectively. This result clearly reflects the increase in the flour protein content due to the treatment. However, the foliar N fertilizer applied at anthesis (T3), only led to an increase in W in the silt loam soil (+26%). In each experiment, the improver wheat fertilized with the granular N fertilization at heading fitted the quality requirements for this market class (W > 300 J  $10^{-4}$  according to the synthetic quality index of Italian classification, Foca et al., 2007), but this qualitative level was never reached for the N foliar application at anthesis.

As far as the P/L rate is concerned, on average, treatment T2 reduced this parameter, compared to T1, by 20 and 17% in the sandy loam and silt loam soils, respectively; while the T3 treatment only reduced this parameter in the silt loam soil in the 2008–2009 growing season (–29%). The variation in P/L was mainly due to the remarkable increase in extensibility (L) (Table 6). As far as the farinograph parameters are concerned, the soil N fertilization at heading (T2) increased water absorption and reduced the degree of softening, in comparison to the T1 control, with the same trend being observed in both environments, while it had a greater influence on the development time in the sandy loam soil. Stability were barely affected by T2, with the exception of silt loam soil site in the 2008–2009 period.

The foliar N fertilizer applied at anthesis (T3) did not generally affect the farinograph parameters, with the exceptions of water absorption and degree of softening at the silt loam soil site and development time at the sandy loam soil site, both in the 2009–2010 growing season. The low stability, and therefore the higher degree of softening observed at this site in the 2008–2009 growing season for the T1 and T3 treatments can be attributed to the very low protein content of the flour observed in those samples.

The ANOVA did not show any significant effect of the late-season N fertilization on the gluten index in any of the experiments conducted on durum wheat, while a significant effect of cultivars was reported for this parameter (Table 5). Only in the experiments conducted in the sandy loam soil, did the late-season N fertilization significantly affect the percentage of YB (P < 0.001). In both growing seasons, the granular N fertilization at heading led to a reduction in YB, while no significant differences were observed for the foliar N fertilizer.

The gluten index values and YB percentage of cultivar Saragolla were higher than those of cultivar Dylan, but the interactions between the late-season N fertilization and the cultivars were never significant for either parameter.

#### **DISCUSSION**

The data of this research have confirmed that, as far as the N management of improver common wheat and durum wheat is concerned, late-season N application is essential to reach the established quality requirements of these crops in the temperate environments. In all of the compared conditions, a granular N fertilizer soil-applied at the beginning of heading was clearly able to prolong green flag leaf area duration during the grain-filling stages and, consistently, increase the GPC of both common and durum wheat, although it was not able to determine any advantage in yield for the crops. These results are in agreement with other studies on common wheat (Johansson et al., 2004; Kratochvil et al., 2005; Zebarth et al., 2007) and on durum wheat (Garrido-Lestache et al.,

Table. 6. Effect of late-season N applications on the alveographic and farinographic parameters†; field experiments conducted in Northwest Italy in the 2008 to 2010 period.

							ph	Brabender farinograph			
Soil	Growing season	Late season N	Flour protein Content	Р	L	P/L	W	Water absorption	Development time	Stability	Degree of softening
			%	n	nm ——		J 10 <sup>-4</sup>	%	min -		BU
Sandy Ioam (Cigliano)	2008-2009	ΤI	11.3	83	89	0.93	290	57.2	1.5	19.0	20
		T2	13.4	91	116	0.78	409	59.6	2.8	18.7	0
		Т3	11.3	83	81	1.02	269	57.7	1.5	19.2	27
	2009–2010	TI	11.6	96	75	1.28	312	56.0	1.5	19.2	35
		T2	13.3	97	100	0.97	408	57.3	2.4	18.9	0
		Т3	11.5	98	63	1.56	266	55.8	2.1	19.0	29
Silt loam (Poirino)	2008–2009	TI	9.5	87	56	1.55	209	56.4	1.4	2.0	73
		T2	11.7	99	82	1.21	332	58.7	1.6	19.0	20
		Т3	10.0	91	83	1.10	297	56.9	1.6	2.1	66
	2009–2010	TI	11.6	97	67	1.45	270	56.3	1.6	19.1	18
		T2	12.9	106	83	1.28	367	57.6	2.0	18.9	0
		Т3	12.2	105	67	1.57	298	57.2	1.6	19.1	3

<sup>†</sup> The reported data refer to the analysis conducted on merged samples of cultivar Bologna, resulting from kernel mixing of the four replications.

2005; Orcen et al., 2013; Ottman et al., 2000) performed in temperate areas, in which it has been reported that N granular late-season soil application increased GPC consistently within different growing seasons.

Moreover, the granular soil N late-season distribution significantly increased TW and grain hardness of common wheat, as a probable consequence of the increase in GPC, but did not lead to any significant differences for durum wheat. Varga and Svečnjak (2006) reported increased kernel weight for late-season N, but only when the plants had been supplied with below optimum N during the prior growth stages, while the increase in TW after late-season N application was always significant. On the other hand, Ruske et al. (2003) observed a linear increase in TW when extra N was applied as foliar urea at anthesis, but not for the late soil application of ammonium nitrate. Ottman et al. (2000) reported that N soil application at anthesis significantly increased TW and TKW in durum wheat. However, a lack of a consistent response of kernel hardness to late-season N on hard red wheat was reported by Altman et al. (1983).

The late-season N application has provided clear benefits to the rheological parameters of common improver wheat, thus confirming previous results (Ayoub et al., 1994; Guttieri et al., 2005; Wuest and Cassman, 1992). According to Borghi et al. (1995) and López-Bellido et al. (2001), as N fertilizer rates increase, a rise in GPC and a corresponding improvement in W and the P/L ratio can be observed. On the other hand, Garrido-Lestache et al. (2004) reported that late-season N application in a semiarid region only affected GPC and the W index, while it did not lead to any change in P/L.

As far as the farinograph parameters is concerned, the impact of N on water absorption and dough development time was consistent with the effect observed for GPC, according to Kharel et al. (2011). The degree of softening decreased with N increasing, while, in disagreement with findings reported by these authors, the effect of late-season N on dough stability was limited.

As far as durum wheat are concerned, a granular N late-season application may reduce YB in the growing season when this defect appears. Poor N availability is considered to be the most critical factor that can influence the presence of YB (Samson et al., 2005). The recorded data are in agreement with those of Gianibelli et al. (1990), who reported that a late application of 15 to 30 kg N ha $^{-1}$ , from heading to postanthesis, decreased the percentage of YB in durum from 80 to 7%, depending on the rate of application and the growth stage.

The present study has reported no differences in the durum wheat gluten index, in function of late-season N fertilization, thus confirming data reported by Garrido-Lestache et al. (2004) concerning common improver wheat. However, Varga and Svečnjak (2006) reported a small, but significant decline in the gluten index of common wheat following a late-season urea treatment.

The N foliar fertilizer provide some benefits, in terms of GPC and other quality requirements, for both common and durum wheat, although its efficacy seems to be variable. A significant positive effect of foliar N applications on increasing the GPC of common and durum wheat has been reported for several irrigated or high rainfall systems (Altman et al., 1983; Blandino and Reyneri, 2009; Ruske et al., 2004; Tea et al., 2007; Varga and Svečnjak, 2006) and in low

Table 7. Costs, net incomes, and return of investment (ROI) for the compared late-season N application strategies, considering different common and durum grain yield levels.

Economic parameters	Factor	T2	T3
		——\$ ha	a <sup>-1</sup>
Late-season fertilization cost†	Application	40	0‡
	N fertilizer	65	37
	Total	105	37
Net incomes§	Grain yield 3 t ha <sup>-l</sup>	10	78
	Grain yield 7 t ha <sup>-l</sup>	163	231
ROI¶	Grain yield 3 t ha <sup>-l</sup>	0.1	2.1
	Grain yield 7 t ha <sup>-l</sup>	1.6	6.2

<sup>†</sup> According to survey conducted in North Italy by Mancuso (2013).

rainfall environments (Bly and Woodard, 2003; Orcen et al., 2013; Woolfolk et al., 2002). An effect of foliar N fertilizer applications on grain yield has been reported in a few cases, although decreased yields following late urea spraying have been observed in other cases (Barraclough and Haynes, 1995; Peltonen et al., 1991). Other experiments on durum wheat conducted in growing areas with different climatic conditions have reported negligible effects of late-season N foliar application on GPC (Abad et al., 2004; Blandino et al., 2009; Garrido-Lestache et al., 2005). Foliar application is less effective at increasing the grain N content when large amounts of N have previously been applied (Gooding and Davies, 1992). Varga and Svečnjak (2006) reported that the benefits of late season foliar applications are influenced by both the cultivar and the plant N status.

Furthermore, in the pedo-climatic conditions considered in this experiment, the strategy to apply a N foliar fertilizer at anthesis, at a commercial rate that is able to keep leaves safe from any leaf burns and that can be applied in NVZ, has resulted in clear lower improvement in GPC and the quality parameters of these end-use market categories, than the soil distribution of a granular fertilizer at a higher N rate. Moreover, the foliar N fertilizer at anthesis led to a less stable breador pasta-making quality enhancement, that seem to be related to the environmental conditions.

The results reported in the present work suggest that foliar N application may be more effective in finer textured soils with higher CEC, than in shallower and coarser-textured soil, characterized by a probable higher nitrate leaching during the vegetative stages. This result was also found by Gooding et al. (2005), who reported that the effect of foliar urea on GPC is closely dependant on the N available from other sources.

The duration of the ripening stages could be another possible interpretation of the variable effectiveness of N foliar fertilizer to enhance grain quality in improver common and durum wheat. In Cigliano site, characterized in both growing season by higher GDDs from flowering to the harvest, and, therefore, by a faster ripening, the foliar N application did not lead to any grain quality enhancements, that otherwise have been observed in Poirino site, where the grain maturation was more gradual. Crops with higher stay green, that is the capacity to retain green leaf areas for longer, have shown less decline in the

<sup>‡</sup> Considering the foliar N fertilizer tank mixed with a fungicide and assigning the application cost all to the disease control treatment applied at anthesis.

<sup>§</sup> Considering a premium price of 15 and 8% and an average price of 255 and 473 \$  $t^{-1}$  for common and durum wheat, respectively.

<sup>¶</sup> ROI = (gain of investment – cost of investment)/cost of investment.

N uptake during ripening (Borrell et al., 2001). As Pepler et al. (2005) have reported, the longevity of flag leaf is closely correlated to the accumulation of proteins in the grain. In previous research, we found that the addition of foliar feed, containing macro- and micronutrients, to a fungicide program at anthesis significantly increased green flag leaf area duration, maintained canopy longevity during grain filling and increased the rheological properties of the derived flour (Blandino and Reyneri, 2009). Delaying leaf senescence, which leads to extended photosynthesis, should provide more carbohydrates to roots for prolonged N uptake (Dreccer, 2006). However, in the present work, although the soil N application significantly affect the flag leaf greenness in any growing season and sites, the effect of foliar N application on this parameter was not consistent in the different environmental conditions analyzed.

In spite of the minor effects that have been observed, foliar applications of N may be beneficial under certain circumstances. Foliar N fertilizers could be an alternative strategy to top-dressed soil fertilizer in non-irrigated wheat areas, characterized by infrequent and limited rainfall from wheat heading to the end of grain filling. In these conditions, N soil application can be less effective, since adequate moist soil surface on which the N can dissolve is lacking and from which it can move deep enough to be taken up by the roots (Brown et al., 2005).

As far as the economic analysis is concerned, a simple comparison between the compared late-season N fertilization strategies is reported in Table 7. The foliar N fertilizer (T3), if applied tank mixed with a fungicide at anthesis, is less expensive and can lead to a greater economic benefit and return on investment than the granular top-dressed soil application (T2). Moreover, the economic convenience of the granular N soil fertilization depends clearly by the wheat yield level and it is inconsistent at a 3 t ha<sup>-1</sup> grain yield.

In conclusion, late-season N fertilization has been confirmed to play a key role in obtaining common improver and durum wheat grains with high GPC, which are able to satisfy the qualitative requirements of these market categories. This research clearly highlight that in all the considered environmental conditions the distribution of soil granular N at heading at a 40 kg N ha<sup>-1</sup> rate clearly led to constant quality improvements, considering several parameters, for both of the compared crops. Moreover, the research has indicated that the strategy to apply a foliar N fertilizer at anthesis at a lower rate led to a less effective and stable bread- or pasta-making quality enhancement, although it has a greater cost effectiveness. The foliar fertilizer, rather than a direct effect on plant N nutrition, could probably play only an indirect effect on maintaining a higher postanthesis N uptake. Although this effect depends to a greater extent on the environmental conditions, this strategies could contribute to a positively enhance GPC in soils with a probable lower nitrate leaching during the vegetative stages.

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