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Sustainable and Profitable Nitrogen Fertilization Management of Potato

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Abstract: Nitrogen fertilization is indispensable to improving potato crop productivity, but there is a need to manage it suitably by looking at environmental sustainability. In a three-season experiment, we studied the effects of five nitrogen (N) fertilization rates: 0 (N0), 100 (N100), 200 (N200), 300 (N300) and 400 (N400) kg N ha^{-1} on crop N uptake, apparent nitrogen recovery efficiency (ANRE), tuber yield, nitrogen use efficiency (NUE), nitrogen uptake efficiency (NUpE), nitrogen utilization efficiency (NUtE) and agronomic nitrogen use efficiency (AgNUE) of five different potato cultivars: Daytona, Ninfa, Rubino, Sieglinde and Spunta. The economically optimum N fertilizer rates (EONFR) were also calculated. In seasons with high soil nitrogen availability for the crop (about 85 kg ha^{-1} of N), tuber yield increased only up to N100 and ANRE was about 50%; in seasons with medium (from 50 to 60 kg ha^{-1} of N) soil N availability, tuber yield increased up to N200 and ANRE was about 45%. Rubino and Sieglinde (early cultivars) responded for tuber yield only up to N100; Daytona, Ninfa, Spunta (late cultivars) up to N200, showing the highest values of NUE, NUpE, NUtE and AgNUE at N100. EONFR ranged from 176 to 268 kg ha^{-1} in relation to cultivar and season, but the reduction by 50% led to a tuber yield decrease of only around 16%. The adoption of cultivars characterized by high AgNUE at a low N rate and a soil nitrate test prior to planting, are effective tools to achieve a more sustainable and cost-effective nitrogen fertilization management.

Keywords: potato; nitrogen fertilization; environmental sustainability; cost-effective; nitrogen use efficiency; tuber yield; EONFR

1. Introduction

Potato is a very important crop in the Mediterranean basin, occupying an overall area of a little less than one million ha and producing 30 million tons of tubers [1]. In several countries such as Tunisia, Morocco, Egypt, Cyprus, Israel, Lebanon, Turkey, Spain and in southern Italy, potatoes are not grown in the usual cycle (spring–summer) owing to the high temperatures and considerable demand for irrigation water, but are largely grown in two offseason crops for early production: Winter–spring (planting from December to January and harvesting from March to early June) [2], and summer–autumn (planting in early September and harvesting from November to the end of January). Early potatoes, defined as "potatoes harvested before they are completely mature, marketed immediately after harvesting and whose skin can easily be removed without peeling" (United Nations Economic Commission for Europe of Geneva, Fresh Fruit and Vegetables-30/2001), are highly appreciated and are mainly exported to northern European countries, with considerable profit [3]. The substantial commercial value of the product and the intensive use of the land prompt farmers to supplement the potato crop with water and nutrients, which have undoubtedly been responsible for increased early potato yields in recent decades. As a consequence of low nitrogen (N) reserves and high mineralization potential

in Mediterranean soils [4], N fertilization is considered indispensable to improve crop productivity. Indeed, N application has a substantial effect on the leaf area index (LAI) of potatoes by increasing both the rate of leaf expansion and the number of emerging leaves, and directly influences seasonal patterns of photon interception and crop production [5,6]. Because of the central role of this macronutrient in determining crop growth and yield capability, N fertilization of the early potato cultivation in the Mediterranean basin is excessive and often even irrational, with N rates higher than 600–700 kg ha^{-1} frequently being applied [7]. These rates are far greater than the usual crop N uptake, which for a tuber yield of about 20 t ha^{-1} is equal to about 100 kg ha^{-1} of N [7]. The excess nitrogen (N) not taken up by the crop remains in the soil profile and may be subject to losses by denitrification, volatilization, surface runoff and leaching to the groundwater, resulting in pollution of the environment [8]. This is favored by the high amounts of irrigation water applied, low efficiency of irrigation methods such as furrow or sprinkler [9] and by light-textured soils [10], common in early potato cultivation. The risk of pollution, as well as the fact that producing mineral N fertilizer is highly demanding in terms of fossil fuels, has increased the urgency for environmental care [11]. The Nitrate Directive 91/676/EEC [12] and the Water Framework Directive 2000/60/EC [13] are implementing a reduction in N supply to crops in Europe. The focus of agronomic research has therefore shifted from finding the optimum rate of input for maximizing tuber yield to how to make best use of the permitted maximum amount of the external supply of N [6]. Environmental losses of N from potato production systems are frequently high despite improvement in fertilizer N management practices. One approach to reducing environmental losses of N is to increase the nitrogen use efficiency (NUE) of the crop. Potato is characterized by a relatively low NUE ranging between 50% and 60% [14], due to it having a naturally shallow and poorly developed root system which is less efficient in taking up N than other crops such as wheat, maize or sugar beet [15]. On the other hand, with exaggerated N fertilization rates, the profit for the producer will also drop, as fertilizers are becoming more expensive [11]. Selection or identification of the N fertilizer rate is one of the most basic, yet most important decisions in managing N fertilizer [16]. For farmers, it has become more important to manage N fertilization in terms of providing a cost-effective yield, even if not necessarily the maximum possible yield, which can reduce environmental impact at the same time. As the N crop response is genotype-dependent [14], it would be useful to have this information on selected cultivars that may differ for biological, morphological and productive traits. With the exception of a few contributions [17–19], each investigating the effects of the nitrogen fertilization rate on the fate of N fertilizer, N uptake capacity and tuber yield in the Mediterranean environment, on one sole cultivar and for no more than two years, no attempts have focused to date on NUE and on defining economically optimum N fertilizer rates for early potato production. The goal of this work was, over a three-season period, (i) to evaluate the effects of different nitrogen fertilization rates on N uptake, tuber yield, nitrogen efficiency and ii) to determine the economically optimum N fertilizer rates in five different genotypes to achieve a more sustainable and cost-effective nitrogen fertilization management of early potato crops in a Mediterranean environment.

2. Materials and Methods

2.1. Site, Climate and Soil

Experiments were conducted in 2010, 2011 (season I and II, respectively) and 2014 (season III) at our experimental field, with a wheat pre-crop two years before, on the coastal plain, south of Siracusa (37°03′ N, 15°18′ E, 15 m a.s.l.), a typical area for potato cultivation in Sicily (South Italy). The climate is semi-arid Mediterranean, with mild winters, and commonly rainless springs. Frost occurrence is virtually unknown (only two events in 30 years). During the potato crop season for early production (from December–January to May), the mean maximum day temperatures and the mean minimum night temperatures of the 30-year period 1977–2006 were 15.4 and 7.1 °C in January, 16.2 and 7.6 °C in February, 17.7 and 8.8 °C in March, 20.2 and 10.9 °C in April, 24.3 and 14.4 °C in May, respectively. Rainfall over the same period averages about 180 mm (Figure 1). In the three seasons of the experiment,

we used three adjoining plots in the same field. The soil, moderately deep, was classified as Calcixerollic Xerochrepts on the basis of the USDA Soil Taxonomy Classification [20]. At the start of the experiments, the soil characteristics analyzed in our laboratory were as follows: Sand (41%), silt (30%), clay (29%), limestone (4%), pH (7.9), organic matter (2.1%), total N (1.6$\%_0$), assimilable P_2O_5 (46 ppm), exchangeable K_2O (414 ppm).
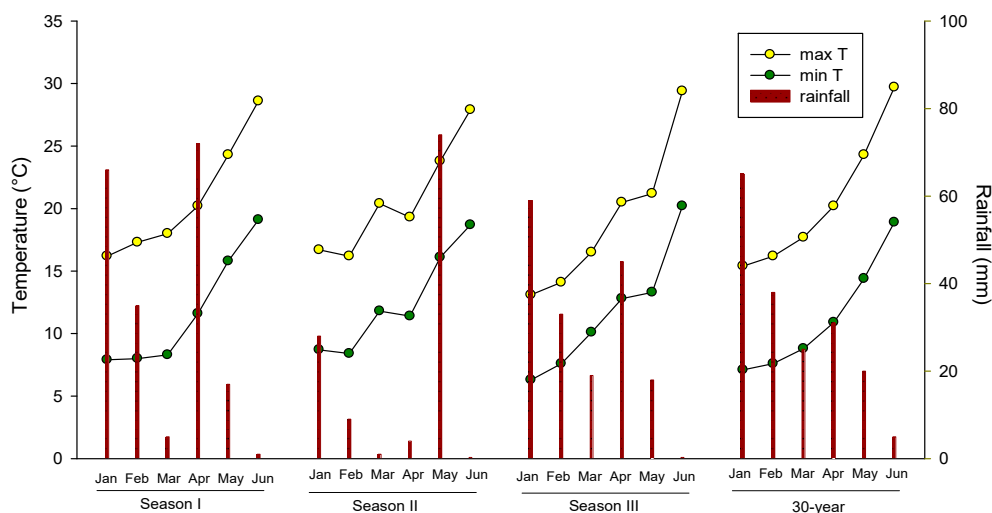
Figure 1. Average monthly maximum and minimum air temperatures and total monthly rainfall for the three seasons and 30-year period.

2.2. Experimental Design, Plant Material and Management Practices

The experiment (seasons I and II) was arranged in a randomized split-plot design with four replications including five nitrogen rates (0, 100, 200, 300, and 400 kg ha^{-1} referred after as N0, N100, N200, N300 and N400) as main plots and four cultivars of potato (*Solanum tuberosum* L.), e.g., Spunta, Sieglinde, Daytona, and Ninfa as sub-plots. In season III, the experiment was arranged in a split-plot design with four replications including five nitrogen rates (N0, N100, N200, N300 and N400) as main plots and two cultivars of potato (Rubino and Ninfa) as sub-plots. The five cultivars (Spunta, Sieglinde, Daytona, Ninfa and Rubino) utilized in this research differ for their morphological, biological, physiological and productive characteristics. Spunta and Sieglinde are widely cultivated in the Mediterranean region. Spunta is an early-medium ripening ware potato with long, regular, and very large tubers; plants produce few erect and vigorous stems and are well adapted to the Mediterranean climate, where they produce a high tuber yield [21]. Sieglinde is a firm flesh early cultivar with oblong, regular, and moderate-sized tubers; plants produce numerous stems of medium height, semi-erect and are moderately vigorous; they usually develop only limited biomass and deliver low tuber yield [2]. Daytona, Ninfa and Rubino are Italian cultivars bred within the Italian project "Breeding of Potato": Daytona was bred by Agenzia per la Sperimentazione Tecnologica e la Ricerca Agroambientale (ASTRA)—Innovation and Development (ex Mario Neri), Imola (Bologna), Ninfa and Rubino by CREA—Research Centre for Cereal and Industrial Crops, Bologna [22]. Daytona is a cultivar of medium to late maturity with short, oval, and regular tubers; stems are of medium size. Ninfa is a cultivar of medium to late maturity, with oblong, regular, and very large tubers; plants produce fairly tall and erect stems and provide marketable tuber yields superior to those of commercial cultivars frequently cultivated in southern Italy. Rubino is an early cultivar with oval and moderate-sized tubers; it was selected for earliness and suitability to early production. Whole virus-free seed-tubers were planted on February 3 (season I), on January 29 (season II) and on January 28 (season III). Plants emerged between 30 and 40 days after planting (DAP). In all experiments, the sub-plot size was 4.2 × 4.2 m, with 84 plants and consisted of six rows; tubers were planted at 0.3 intervals, in rows 0.7 m apart (equivalent to a planting density of 4.76 plants m^{-2}). In the three seasons, tillage consisted

of a 40 cm depth ploughing followed by harrowing in October; at planting 100 kg ha^{-1} of P_2O_5 (as mineral superphosphate) and 150 kg ha^{-1} of K_2O (as potassium sulphate) were applied, whereas 50% of nitrogen (as ammonium nitrate) was supplied at complete crop emergence and the remaining 50% three weeks after as top dressing. Chlorpyrifos (30 kg ha^{-1}) was applied before planting; other standard crop management was applied, involving post-emergence weeding with linuron and pest control when needed. Crop water requirements were completely satisfied by drip irrigation, supplying 100% of crop maximum evapotranspiration, when the accumulated daily evaporation measured by class A pan evaporimeter reached 30–40 mm. Over the crop cycle, 197 (season I), 210 (season II) and 170 mm (season III) irrigation water were applied.

2.3. Data Collection and Calculations

2.3.1. SPAD Measurements

Leaf SPAD absorbance (correlated to chlorophyll content) was measured in the field using a portable Chl meter (SPAD 502, Minolta Camera, Osaka, Japan). Measurements were made on the distal leaflet of the youngest fully expanded leaf (usually the third or fourth leaf from the apex) between 11:00 and 13:00 (local solar time). Triplicate readings were taken from fully sun-exposed leaflets of 4 potato plants randomly sampled in four central rows of each sub-plot [23]. Between the 5th–6th leaf appearance and beginning of plant senescence, ten measurements were taken in season I, seven in season II and four in season III.

2.3.2. Plant Weight and Tuber Yield

When about 70% of leaves were dry (126, 121 and 120 DAP in season I, II and III, respectively), plants from central rows of each subplot were hand collected by removing an undisturbed soil sample. Plants were separated into aboveground biomass (stem + leaves), roots + stolons and tubers; roots, stolons and tubers were washed in gently running water. Tubers were classified in marketable (unitary weight > 20 g) and unmarketable (unitary weight < 20 g). All plant parts (marketable and unmarketable tubers, aboveground biomass and roots) were weighed separately to measure fresh weight. Marketable tubers were utilized to determine tuber yield. Three samples of about 50 g of all plant parts for each plot were oven-dried at 105 °C until constant weight and weighed to determine dry matter content.

2.3.3. Economically Optimum N Fertilizer Rate

To predict the economically optimum N fertilizer rates (EONFR), a quadratic equation model (SigmaPlot 11, Systat Software Inc.) described by Fontes et al. [24] and Belanger et al. [25] was utilized:

$$Y = b_0 + b_1N + b_2N^2 \tag{1}$$

where Y is the expected marketable fresh tuber yield expressed in kg ha^{-1}, N is applied fertilizer N expressed in kg ha^{-1}, and b_0, b_1 and b_2 are coefficients that are calculated from the experimental data. The EONFR, defined as the rate of N application where €1 of additional N fertilizer returned €1 of potatoes, was calculated as follows:

$$N_{op} = P - b_1/2b_2 \tag{2}$$

where N_{op} is the economically optimum application rate of fertilizer N expressed in kg ha^{-1}, P is the ratio of the cost of N fertilizer (€ 1.6 kg^{-1} N) to the selling price of potatoes (€ 0.25 kg^{-1} tuber), on the average of 2015 to 2016 [26], b_1 and b_2 as in Equation (1); this analysis assumes that fertilizer N costs are the only variable costs and that all other costs are fixed. According to Neeteson [27], EONFR was adjusted by considering the amount of available N in the soil at planting according to the following formula:

$$N_{op} = P - b_1/2b_2 - 0.7 N_A \tag{3}$$

N_A represents the amount of available N in the 0–0.40 m soil layer at planting, which was the sum of mineral N (NO_3-N and NH_4-N) plus the N released through mineralization during the growing season, plus the N supplied through fertilization [28]. The initial mineral nitrogen content of the soil profile (0–0.40 m) was set at about 1% of total N, determined by the Kjeldahl method. N soil mineralization per month was calculated according to Gariglio et al. [29], from total N content corrected by an N mineralization factor as a function of soil temperature. On the basis of this procedure, the quantity of available mineral nitrogen in the soil for the crop cycle at planting was, regardless of cultivars, about 48 kg ha^{-1} in season I, 84 kg ha^{-1} in season II and 64 kg ha^{-1} in season III. In addition, four reduced rates (90, 75, 50 and 25%) of N_{op} were simulated and the relative yields decrease was calculated from the response quadratic curves.

2.3.4. Determination of Crop Nitrogen Content and Nitrogen Uptake

Nitrogen concentration in roots + stolons, marketable and unmarketable tubers and above-ground biomass was determined for each replication by dried materials collected at harvest, which was finely ground through a mill (IKA, Labortechnick, Staufen, Germany) with a 1.0 mm sieve. Nitrogen was determined by means of the Kjeldahl method (Kjeltec 2300 Auto Analyser; Foss-Tecator, Hilleroed, Denmark) [30]. The N content of each part of the plant was calculated as the product of the measured N concentration and dry weight (DW). Crop nitrogen uptake (CNU) was calculated as the sum of N contents of roots + stolons, marketable and unmarketable tubers and aboveground biomass.

2.3.5. Nitrogen Efficiency Indices

The efficiency of N fertilizer utilization was calculated using the following equation adapted from Vos [6]:

$$ANRE = (N_U - N_0)/N_F \; 100 \tag{4}$$

where ANRE is apparent nitrogen recovery efficiency expressed in %, N_U is the N uptake of the N-fertilized plot, N_0 is the uptake of the N-unfertilized plot (control), N_F is the amount of N applied by fertilization.

The inefficiency of N fertilizer utilization was calculated as follows:

$$NRI = 100 - ANRE \tag{5}$$

where NRI is nitrogen recovery inefficiency expressed in %.

Using the following equations adapted from van Bueren and Struik [8]:

$$NUE = Y_N/N_A \tag{6}$$

where NUE is nitrogen use efficiency expressed as kg ha^{-1} tuber DW kg ha^{-1} N, Y_N is marketable dry tuber yield, N_A as in Equation (3);

$$NUpE = N_U/N_A \tag{7}$$

where NUpE is nitrogen uptake efficiency expressed as kg ha^{-1}N kg^{-1} ha^{-1} N, N_U represents the amount of N uptake by the crop, N_A as in Equation (3);

$$NUtE = Y_N/N_U \tag{8}$$

where NUtE is nitrogen utilization efficiency expressed as kg ha^{-1} tuber DW kg ha^{-1} N, Y_N as in Equation (6), N_U as in Equation (7).

$$AgNUE = Y_N/N_F \tag{9}$$

where AgNUE is agronomic nitrogen use efficiency expressed as kg ha^{-1} tuber DW kg ha^{-1} N, Y_N as in Equation (6), N_F as in Equation (4).

2.4. Meteorological Data

Air temperature, relative humidity and rainfall were monitored during the experiments by a meteorological station (CR 21 data logger, Campbell Scientific, Inc., Utah, U.S.A.) sited at the experimental field. Measurements were made every 30 min.

2.5. Statistical Analysis

Data collected were first submitted to Bartlett's test to check the homoscedasticity, then analyzed using ANOVA [31]. A preliminary statistical analysis done for the same cultivars for season I and II showed a significant ($P = 0.001$) effect of interaction "season x nitrogen rate" for all parameters with the exception of the SPAD reading, indicating that the average response of 4 cultivars to the N rate was different for seasons I and II; the other preliminary statistical analysis made for Ninfa in all three seasons (I, II and III) showed a significant ($P = 0.001$) effect of interaction "season x nitrogen rate" for all parameters indicating that its response to N rate was different for the three seasons. Consequently, we analyzed each season's results separately, based on a factorial combination of "nitrogen rate × cultivar". Means were compared by a Least Significant Difference (LSD) test, when the F-test was significant. Table 1 shows the statistical significance from the analysis of variance for all studied variables separately for each season. CoStat Version 6.003 (CoHort Software, Monterey, CA, USA) was used. Polynomial effects up to the second degree were made where appropriate to define the linear or quadratic trend of N treatments and all studied parameters.

Table 1. Summary of statistical significance from analysis of variance for all studied variables: Crop nitrogen uptake (CNU), apparent nitrogen recovery efficiency (ANRE), nitrogen use efficiency (NUE), nitrogen uptake use efficiency (NUpE), nitrogen utilization use efficiency (NUtE), agronomical nitrogen use efficiency (AgNUE), in the three seasons; df indicates degree of freedom; **, *** indicate significant at $P \leq 0.01$, 0.001, respectively; NS = not significant.

Variable	Source of Variation	df	Season I	Season II	df	Season III
CNU	Nitrogen rate (N)	4	***	***	4	***
	cultivar (C)	3	***	***	1	**
	(N) × (C)	18	***	***	8	***
SPAD readings	Nitrogen rate (N)	4	***	***	4	***
	cultivar (C)	3	***	***	1	***
	(N) × (C)	18	***	***	8	**
ANRE	Nitrogen rate (N)	3	***	***	3	***
	cultivar (C)	3	***	***	1	***
	(N) × (C)	14	**	***	6	**
Tuber yield	Nitrogen rate (N)	4	***	***	4	***
	cultivar (C)	3	***	***	1	**
	(N) × (C)	18	***	**	8	***
NUE	Nitrogen rate (N)	4	***	***	4	***
	cultivar (C)	3	***	***	1	**
	(N) × (C)	18	***	**	8	***
NUpE	Nitrogen rate (N)	4	***	***	4	***
	cultivar (C)	3	***	***	1	NS
	(N) × (C)	18	***	**	8	NS
NUtE	Nitrogen rate (N)	4	***	***	4	**
	cultivar (C)	3	**	***	1	***
	(N) × (C)	18	NS	NS	8	***
AgNUE	Nitrogen rate (N)	3	***	***	3	***
	cultivar (C)	3	***	***	1	***
	(N) × (C)	14	**	**	6	***

2.6. Weather Conditions

The average monthly maximum and minimum temperatures from January to June were similar in the 3 seasons and to the 30-year (1977/2006) average, with the exception of March (Figure 1). In that month in season II, monthly maximum temperatures were 2.4 °C higher than in season I, 3.9 °C higher than season III and 2.7 °C higher than the 30-year average; minimum temperatures were higher by

3.5 °C compared to season I, by 1.7 °C compared to season III and by 3.0 °C compared to the 30-year average. The volume of rainfall from January to June and the distribution was similar in season I (196 mm) and season III (174 mm) and also with respect to the 30-year mean (184 mm); during season II rainfall was lower (116 mm) and was concentrated for about 65% in May, whereas it was absent in March and April (Figure 1).

3. Results

3.1. Crop Nitrogen Uptake, SPAD Readings, ANRE

In N0 plots, CNU, averaged over cultivars, was about 48 (season I), 84 (season II) and 64 (season III) kg ha^{-1} (Table 2). In N-fertilized plots, CNU increased linearly (all cultivars and seasons) and quadratically (all cultivars in season I and in season III) with the increase of the N rate (Table 2). Our results also indicate that CNU in relation to the N rate was cultivar-dependent. In fact, the highest increase in N uptake, increasing from N100 to N400, was found in Spunta (96 and 52 kg ha^{-1}, respectively in season I and II) and in Ninfa (104 and 91 kg ha^{-1} respectively in season I in season II); the lowest in Sieglinde (43 and 24 kg ha^{-1} respectively in season I in season II). In season III (in the same intervals) Rubino showed far less increases of N uptake (7 kg ha^{-1}) than Ninfa (36 kg ha^{-1}) (Table 2).

Table 2. Crop nitrogen uptake and SPAD readings as affected by "nitrogen rate x cultivar" interaction in the three seasons. Relationship tested by regression analysis, between N rate and responses of each variable and cultivar (L = linear, Q = quadratic; *, **, *** indicate significance at $P \leq 0.05$; $P < 0.01$; $P \leq 0.001$.

Season	N Rate	Crop Nitrogen Uptake (kg ha^{-1})				SPAD Readings (units)			
		Spunta	Sieglinde	Daytona	Ninfa	Spunta	Sieglinde	Daytona	Ninfa
I	N0	43.3	47.4	52.2	48.9	36.0	32.9	33.1	34.0
	N100	113.2	111.4	115.8	114.3	40.0	35.1	36.9	36.1
	N200	158.4	137.4	160.4	163.7	41.1	37.1	39.0	38.0
	N300	185.9	156.4	175.1	171.1	44.0	37.0	39.8	38.9
	N400	209.0	154.5	190.0	217.9	45.1	39.1	39.9	39.9
	L	***	***	***	***	***	***	***	
	Q	***	***	***	***			***	**
		LSD inter. ($P \leq 0.05$) **15.9**						**1.3**	
II	N0	74.9	83.5	90.9	88.1	38.6	35.7	36.0	33.3
	N100	116.0	131.3	156.1	133.0	39.3	38.6	40.0	36.7
	N200	107.7	115.3	162.8	145.1	41.2	39.3	40.6	38.9
	N300	166.2	126.8	177.0	159.0	43.0	40.0	41.3	40.0
	N400	168.0	154.9	201.3	198.8	44.6	39.0	40.7	40.7
	L	***	***	***	***	***	*	***	***
	Q						*	**	*
		LSD inter. ($P \leq 0.05$) **27.7**						**1.2**	
III			Rubino	Ninfa				Rubino	Ninfa
	N0		67.1	60.5				32.4	34.8
	N100		106.4	115.2				37.9	39.3
	N200		105.9	128.3				38.4	40.9
	N300		106.6	123.6				40.7	41.8
	N400		113.7	151.0				38.1	41.9
	L		***	***					*
	Q		**	***					
		LSD inter. ($P \leq 0.05$) **9.7**						**1.2**	

Chlorophyll meter readings, measured by SPAD-502, increased with increase of the nitrogen rate (Table 2). Generally, the major increases in SPAD units, rising from N100 to N400, were found in Spunta and Ninfa, and less so in Sieglinde and Rubino, confirming what was found for CNU.

ANRE, which expresses the proportion of N applied taken up by the plants, generally showed higher values in season I (50%) than in season II (28%) and season III (26%) (Table 3).

Table 3. Apparent nitrogen recovery efficiency (ANRE) and tuber yield as affected by "nitrogen rate x cultivar" interaction in the three seasons. Relationship tested by regression analysis, between N rate and responses of each variable and cultivar (L = linear, Q = quadratic; *, **, *** indicate significance at $P \leq 0.05$; $P \leq 0.01$; $P \leq 0.001$.

Season	N Rate	ANRE (%)				Tuber Yield (t ha^{-1})			
		Spunta	Sieglinde	Daytona	Ninfa	Spunta	Sieglinde	Daytona	Ninfa
I	N0	-	-	-	-	14.3	14.5	19.4	20.5
	N100	80	64	64	65	35.9	31.1	35.9	37.0
	N200	62	45	54	57	46.7	35.3	43.5	47.4
	N300	51	36	41	41	48.7	38.1	41.1	46.7
	N400	44	27	34	42	48.5	35.1	39.0	46.2
	L	***	***	***	***	***	***	***	***
	Q					***	***	***	***
	LSD inter. ($P \leq 0.05$) **6.0**					**5.0**			
II	N0	-	-	-	-	35.7	25.3	43.8	40.6
	N100	41	48	65	45	47.7	36.8	50.8	47.5
	N200	30	16	36	28	47.6	33.8	55.6	50.7
	N300	16	14	29	24	53.3	32.8	58.0	48.5
	N400	23	18	28	28	54.6	35.2	61.8	48.2
	L	*	**	***	**	***		**	
	Q	*	**	*	*				*
	LSD inter. ($P \leq 0.05$) **11.2**					**5.4**			
III			Rubino	Ninfa			Rubino	Ninfa	
	N0		-	-			19.7	16.3	
	N100		39	55			28.7	32.9	
	N200		19	34			30.9	43.0	
	N300		13	21			25.0	40.1	
	N400		12	23			28.3	33.6	
	L		***	***			*	***	
	Q		***	***			**	***	
	LSD inter. ($P \leq 0.05$) **3.0 3.7**								

As shown in Table 3, ANRE linearly (all cultivars and seasons) and quadratically (all cultivars in season II and in season III) decreased with the increasing nitrogen rate. In season I and II, the decrease in ANRE from N100 to N400 was more pronounced in Sieglinde (−58% and −62%, respectively in season I and II) than in Spunta (−45% and −44%, respectively), Daytona (−47% and −57%, respectively) and Ninfa (−35% and −38%, respectively); in season III the decrease was more pronounced in Rubino (−69%) than in Ninfa (−58%).

NRI (see Equation (4)), which represents the possible environmental impact, increased dramatically with the increasing N rate reaching N400, regardless of cultivars, and with values of about 63% in season I, 76% in season II and 83% in season III (data not shown).

3.2. Tuber Yield and Economically Optimum N Fertilizer Rate

In unfertilized N$_0$ plots, marketable tuber yield was, averaged over cultivars, 17.2 t ha^{-1} (season I), 36.3 t ha^{-1} (season II) and 18.0 t ha^{-1} (season III) (Table 3). In N fertilized plots, tuber yield increased linearly and quadratically in all cultivars (season I and III) with the increase of the N rate (Table 3). In season II the increase was linear in Spunta and Daytona, whereas it was quadratic in Ninfa (Table 3). Sieglinde (season I and II) and Rubino (season III), responded noticeably only up to N100, whereas Ninfa in the three seasons responded up to N200. Spunta and Daytona in season I responded significantly up to N200, whereas in season II were able to exploit higher doses of N fertilizers (300 kg ha^{-1}).

EONFR for each cultivar and season are reported in Table 4. For cultivar Ninfa, the sole cultivar to be used in all three seasons, EONFR changed substantially over the three seasons, showing a clear increasing trend (from 176 (season II), to 197 (season III), to 254 (season I) kg N ha^{-1}) with decreasing soil N mineral availability reserves (from 84 to 64, to 48 kg N ha^{-1}, respectively).

Table 4. Quadratic equations, the economically optimum nitrogen fertilizer rate (EONFR) and corresponding tuber yields in relation to cultivar and season.

Cultivar	Season	Quadratic Equation	R	EONFR (kg N ha^{-1})	Tuber Yield (t ha^{-1})
Spunta	I	$Y = 15094 + 230.9 \, x - 0.374 \, x^2$	0.995	268	50.1
Sieglinde	I	$Y = 15380 + 164.2 \, x - 0.290 \, x^2$	0.989	241	38.1
Daytona	I	$Y = 20157 + 179.2 \, x - 0.337 \, x^2$	0.986	227	43.5
Ninfa	I	$Y = 20897 + 189.9 \, x - 0.322 \, x^2$	0.993	254	48.3
Ninfa	II	$Y = 41031 + 72.8 \, x - 0.141 \, x^2$	0.963	176	49.4
Ninfa	III	$Y = 16363 + 210.9 \, x - 0.423 \, x^2$	0.995	197	41.5
Rubino	III	$Y = 21034 + 69.2 \, x - 0.139 \, x^2$	0.773	181	29.0

However, a decrease in EONFR led to a much less than proportional tuber yield decrease (Figure 2); applying 90% of EONFR, the yield decreased by only about 2%, applying 75% of EONFR, the yield decreased by only about 6%; applying 50% of EONFR, yield decreased by only about 6% in Ninfa in season II, 10% in Rubino in season III and about 19% on average in the other cultivars.
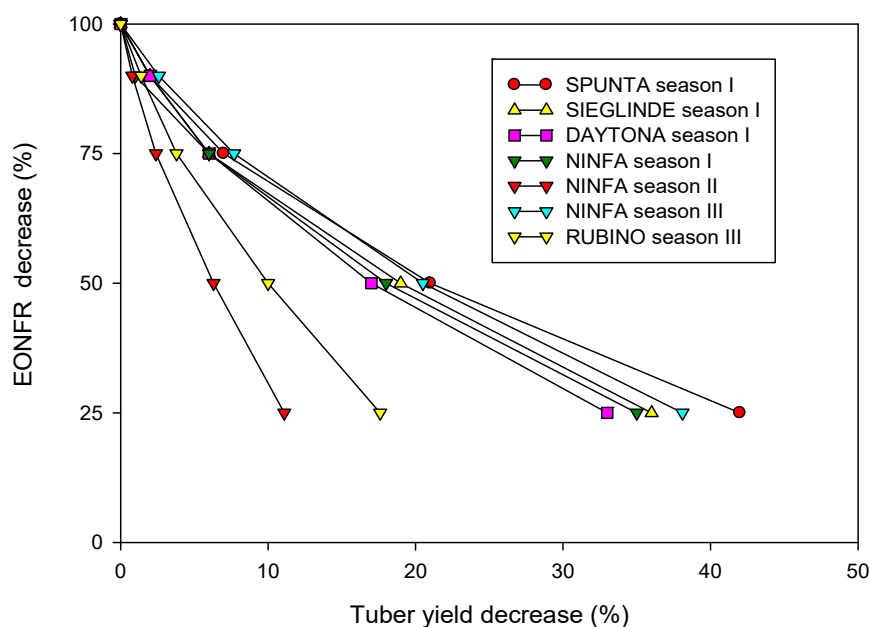
Figure 2. Tuber yield as affected by economically optimum nitrogen fertilization rate in relation to cultivar and season.

3.3. Nitrogen Efficiency Indices

The four nitrogen efficiency indices studied (NUE, NUpE, NUtE, AgNUE) tend to decline linearly and quadratically with increasing N application rates (Table 5; Table 6). The magnitude of this decline was generally genotype-dependent as demonstrated by the significance (nine cases out of 12) of the "nitrogen rate x cultivar" interaction. Sieglinde, compared to Daytona and Ninfa, showed a less evident decrease in NUE with increasing N rates up to N100 (season I) and up to N200 (season II); Rubino, in season III showed a more drastic decrease than Ninfa.

Table 5. NUE (nitrogen use efficiency) and NUpE (nitrogen uptake efficiency) as affected by "nitrogen rate x cultivar" interaction in the three seasons. Relationship tested by regression analysis, between N rate and responses of each variable and cultivar (L = linear, Q = quadratic; *, **, *** indicate significance at $P < 0.05$; $P < 0.01$; $P < 0.001$.

Season	N Rate	NUE (kg Tuber DW kg N^{-1})				NUpE (kg N kg N^{-1})			
		Spunta	Sieglinde	Daytona	Ninfa	Spunta	Sieglinde	Daytona	Ninfa
I	N0	68.2	83.0	112.3	101.8	0.64	0.91	1.00	0.94
	N100	52.5	45.2	57.4	50.3	0.74	0.73	0.76	0.75
	N200	36.4	30.4	41.1	37.0	0.63	0.55	0.64	0.65
	N300	25.3	22.8	25.8	23.4	0.53	0.44	0.50	0.49
	N400	20.9	16.5	20.7	18.6	0.46	0.34	0.42	0.48
	L	***	***	***	***	***	***	***	***
	Q	**	***	***	***	**		*	
	LSD inter. ($P \leq 0.05$)			14.4				0.09	
II	N0	78.0	63.2	111.5	102.6	0.81	0.90	0.98	0.95
	N100	49.6	44.8	62.4	55.9	0.60	0.68	0.81	0.56
	N200	32.2	25.7	42.6	39.6	0.37	0.39	0.56	0.50
	N300	26.8	18.7	30.6	28.0	0.42	0.32	0.45	0.40
	N400	22.4	15.1	24.3	25.1	0.34	0.31	0.41	0.40
	L	***	***	***	***	***	***	***	***
	Q	***	**	***	**	**	**	*	***
	LSD inter. ($P \leq 0.05$)			12.7				0.1	
III				Rubino	Ninfa			Rubino	Ninfa
	N0			51.5	49.8			1.00	0.93
	N100			28.7	36.9			0.63	0.67
	N200			17.8	25.7			0.39	0.47
	N300			11.3	16.3			0.29	0.34
	N400			10.1	12.3			0.24	0.32
	L			***	***			***	***
	Q			***				***	***
	LSD inter. ($P \leq 0.05$) **3.1**								

Table 6. NUtE (nitrogen utilization efficiency) and AgNUE (agronomical nitrogen use efficiency) as affected by the "nitrogen rate x cultivar" interaction in the three seasons. Relationship tested by regression analysis, between N rate and responses of each variable and cultivar (L = linear, Q = quadratic; *, **, *** indicate significance at $P < 0.05$; $P < 0.01$; $P < 0.001$.

Season	N Rate	NUtE (kg Tuber DW kg N^{-1})				AgNUE (kg Tuber DW kg N^{-1})			
		Spunta	Sieglinde	Daytona	Ninfa	Spunta	Sieglinde	Daytona	Ninfa
I	N0	81.5	70.4	87.3	82.1	-	-	-	-
	N100	64.7	56.6	69.2	62.0	73.3	63.1	80.1	70.2
	N200	55.1	53.6	61.5	54.1	43.7	36.3	49.2	44.2
	N300	46.4	49.7	50.4	47.1	28.7	25.8	29.1	26.4
	N400	44.2	47.0	47.9	37.5	23.0	18.1	22.8	20.3
	L	***	***	***	***	***	***	***	***
	Q	***			*	***	***	***	***
	LSD inter. ($P \leq 0.05$)				6.4				
II	N0	100.8	72.0	115.1	108.5	-	-	-	-
	N100	82.4	65.5	77.3	100.9	95.8	86.5	120.4	107.9
	N200	87.1	65.9	77.1	80.3	47.1	37.7	62.3	58.0
	N300	63.5	57.7	67.6	69.1	35.1	24.5	40.1	36.7
	N400	65.6	48.2	59.8	62.2	27.6	18.6	30.0	31.0
	L	**	*	***	***	***	***	***	***
	Q					***	**	**	***
	LSD inter. ($P \leq 0.05$)				11.0				
III				Rubino	Ninfa			Rubino	Ninfa
	N0			48.6	54.3			-	-
	N100			48.1	60.8			47.9	58.9
	N200			45.9	70.1			23.8	33.3
	N300			38.3	67.0			13.8	19.6
	N400			41.2	46.4			11.9	14.1
	L			*				***	***
	Q				***			***	***
	LSD inter. ($P \leq 0.05$) **8.2 3.4**								

Values of AgNUE reported in Table 6, show how in seasons I and II, Daytona proved to be the most efficient in the productive use of nitrogen supplied, while Sieglinde resulted in being the least effective; in season III, Rubino showed lower values of AgNUE than Ninfa at all N rates.

4. Discussion

Nitrogen fertilization has proved an effective means of improving tuber yield in the Mediterranean environment. Although yields responded positively to N application, there was no significant and consistent response of potato crops to varying nitrogen levels above 100 or to the maximum of 200 kg ha^{-1} of the N rate, depending on season and cultivar. In similar Mediterranean environments [7,18], potato yields increased with increasing nitrogen rate up to 120 kg ha^{-1}, but did not change further with higher rates. Even if increasing the nitrogen rate further does not lead to greater tuber yield, it did result in an increase of crop nitrogen uptake and chlorophyll meter readings, measured by SPAD-502, which are considered a promising tool to assess the N status of the potato crop [23,32]. The increase in crop nitrogen uptake was linear in agreement with Vos [6] and/or quadratic as highlighted by Darwish et al. [18] and Badr et al. [19]. In this research, values of crop nitrogen uptake were found to be lower than those in other Mediterranean environments at equal doses of N fertilizers applied [18,19]. This is mainly attributable to the fact that we distributed nitrogen top-dressed in the solid state (as is usually applied), whereas these researchers used fertigation, which is known to enhance N recovery and N use efficiency [17,18]. Increasing nitrogen rates resulted in a marked decrease of nitrogen recovery efficiency (ANRE), in agreement with other authors [18,19,33] and in a decrease of all nitrogen efficiency indices studied, confirming the trends reported by literature [8,16,18,19,24,34]. On average, passing from N100 to N200, the yield increased from 38.4 to 43.4 t ha^{-1}, while the ANRE decreased from 57% to 38%; by further augmenting the N fertilizer the yield remained constant, whereas the ANRE dropped drastically until reaching the maximum dose studied (N400) values of about 28%. This means that applying N400, which is very close to the conventional N fertilizer application dose, a significant amount of fertilizer, about 290 kg ha^{-1} of N, remained not up-taken by the crop and unused in the soil. Only a small part of the N given in excess carries over to the succeeding crops, whereas most of fertilizer N applied to potato is presumably lost over summer by volatilization (N_2O and NH_3) and in autumn, when rainfall exceeds evapotranspiration, by leaching of NO_3 and becomes a risk especially for groundwater and watercourses. ANRE values of about 60% found in N100 plots are in agreement with those found in semi-arid regions [18] and in temperate areas [33], while in more arid areas, such as Turkey and Jordan, values < 40% were found [10,17]. Greenwood and Drycott [34] attributed lower values of ANRE in potato crop, compared to other crops like cereals and grasses, to its lower root density, which causes some of the N fertilizer applied to potato to be remote from the roots for a considerable time before being absorbed. The theoretical economically optimum N fertilizer rates were quite high, ranging from 176 to 268 kg ha^{-1} of N in relation to cultivar and season. This is mainly due to the use of the quadratic model for the calculation of EONFR, which tends to overestimate them [35]. However, the quadratic model was chosen because it proved the most suitable for predicting EONFR because it minimizes the risks of potential economic losses in relation to the cost of the fertilizer and sell price of potatoes [25]. Nonetheless, considering that a decrease in EONFR led to a much less than proportional yield decrease; for example, applying 50% of EONFR, yield decreased by only about 16%, indicating how also from a cost viewpoint, it is possible to reduce currently excessive applications. The response of crop to N fertilization rates was season-dependent. Differences among seasons may largely be attributable to weather conditions, in particular to rainfall occurring before planting. Indeed, very high rainfall in the three months before planting in season I (with a peak of 388 mm in November) and in season III (peak of 350 mm in September) were recorded compared to season II, in which the rains did not exceed 70 mm monthly (data not shown). The high and concentrated rains in the autumn of season I and III probably favored NO_3 leaching, leaving less N availability for the crop in the soil (48 and 64 kg ha^{-1} in season I and III compared to 84 kg ha^{-1} of N in season II). This significantly affected production response of the crop. In unfertilized plots, tuber yields

were in fact only about 17.0 t ha^{-1} in season I, 18.0 t ha^{-1} in season III compared to 36.0 t ha^{-1} in season II, in agreement with Greenwood and Draycott [34] and Rodrigues et al. [36], who found crop response to N rate depends on soil N availability at preplant. Moreover, in fertilized plots, tuber yield response to nitrogen fertilization was up to N200 in season I and III and only up to N100 in season II. In addition, the agronomic response to nitrogen seems to depend largely on soil nitrogen availability, as demonstrated by higher values in all cultivars of the agronomic use efficiency in season II, compared to season I and season III. The EONFR for cultivar Ninfa, the sole cultivar to be used in all three seasons, showed a clearly increasing trend with decreasing soil N mineral availability reserves. Therefore, in the Mediterranean environment, characterized by variability in the amount and distribution of autumn rains, nitrogen fertilization should be commensurate to rainfall. This also suggests the importance of carrying out the soil N concentration test before potato planting, already recommended in some cases to predict the fertilizer N rate in other crops [16]. Our results also indicate that crop response in relation to N rate was cultivar-dependent. Generally, the early cultivars Sieglinde (season I and II) and Rubino (season III), compared to the medium or late cultivars Spunta, Daytona and Ninfa, with an increasing N rate from N100 to N400 showed less increase in plant N uptake and SPAD units and a more pronounced decrease in nitrogen recovery efficiency. Furthermore, they showed less ability to use the soil N available (N residual + N fertilizers) for production of tuber dry matter (NUE), due to both their generally lower removal efficiency of available N (NUpE) and by less efficiency of N taken up to produce yield (NUtE). The lower values of NUpE of the early compared to the late cultivars may be due to the smaller size of the root system [37], whereas the lower NUtE values can be attributed to the shorter crop cycle, lower canopy size (lower % soil coverage), and lower photosynthesis activity [8]. Our previous research has shown how Sieglinde usually develops only limited biomass and delivers low tuber yield [2]. The specific literature reports that differences in NUE among potato cultivars are attributed to their different earliness [14,16,38]. Sieglinde and Rubino also proved less efficient in the productive use of nitrogen supplied than Spunta, Daytona and Ninfa, and responded for tuber yield markedly only up to N100, whereas Spunta, Daytona and Ninfa, were able to exploit higher doses of N fertilizers up to N300 in some cases. Van Bueren and Struik [8], studying a large set of cultivars, found that the majority of genotypes performing well under low N and showing a good response to N were late. Therefore, in consideration of the fact that crop nitrogen uptake generally grows only up to N100 in early cultivars and up to N400 in late cultivars, these last ones have luxury consumption of nitrogen fertilizers in plots supplied with higher N rate (> 200 kg ha^{-1}).

5. Conclusions

This experiment demonstrated that the potato crop, despite the variability between the seasons and the cultivars, benefited only from up to 100, and at most up to 200 kg ha^{-1}, of nitrogen, namely much lower rates than those usually supplied. The variability of the response to N supplied (100 or 200 kg ha^{-1}) found between the years seems to be due to variations in soil available N for the crop over the years. This suggests it is advisable to carry out the soil N concentration test before planting. The early cultivars like Rubino and Sieglinde responded well only up to 100 kg ha^{-1} of nitrogen, the late cultivars Spunta, Daytona and Ninfa up to 200 kg ha^{-1} of nitrogen, also showing, under low N, high agronomic use efficiency. Furthermore, the theoretical economically optimum N fertilizer rates, ranging from 176 to 268 kg ha^{-1} of N in relation to cultivar and season, could be halved without suffering any major yield reduction. Our results can be used to optimize and thus reduce nitrogen fertilization, thereby making savings for the farmer and ensuring a more environmentally-friendly crop in the Mediterranean.

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