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Effect of organic and inorganic fertilizers applied during successive crop seasons on growth and nitrate accumulation in lettuce

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Abstract

A romaine-type lettuce (*Lactuca sativa* L.) cv. Corsica was cultivated during three successive crop seasons (late-spring, late-autumn and late-winter) in the same soil of an experimental greenhouse in S.W. Peloponnese, Greece. Seven long-term fertilization treatments were tested for their effect on plant growth and nitrate concentration in the external lettuce leaves. Treatments included: three different doses of organic fertilization (composted sheep manure) applied at the start of each crop season, three different doses of inorganic N fertilization applied *via* fertigation during each crop season, and a control treatment in which no fertilizer was applied. A drip irrigation system was used to water all plants. The highest nitrate levels were observed in the medium and maximum inorganic fertilization treatments (572–664 mg kg^{-1}) in all crop seasons. They were significantly higher compared to the respective organic fertilization treatments (253–435 mg kg^{-1}) and all other fertilization treatments (148–435 mg kg^{-1}). Crop season affected lettuce growth more than nitrate accumulation in the lettuce leaves: lettuce biomass production was the smallest and most uniform in the late-autumn season and did not respond to the fertilization treatments tested (ranging from 409 to 439 g $plant^{-1}$), while in the late-spring season biomass production was the highest and most variable (561–841 g $plant^{-1}$), it correlated with nitrate concentration in the leaves and in the medium and maximum inorganic fertilizer doses it significantly exceeded production from all other fertilization treatments (827–841 g $plant^{-1}$). Following the three crop seasons the residual availability of N, P and K was clearly enhanced in the soil receiving the organic compared to the inorganic fertilization. Nitrate concentration in lettuce leaves was far below the upper limits set by the European Commission in all fertilization treatments throughout the three crop seasons, a result attributed mainly to the sufficient level of light intensity and duration throughout the year in Southern Greece.
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1. Introduction

In the temperate zone regions lettuce is cultivated throughout the year, but production and quality characteristics benefit from cool weather and high light intensity. The concentration of nitrates in the edible leaves of lettuce are regulated by the European Commission Regulation No 563/2002 which has set upper limits in order to protect consumers from potential toxicological risks following the consumption of nitrate-rich foods (Maynard et al., 1976; Walker, 1990; Bruning-Fann and Kaneene, 1993). Recent studies however, report on beneficial effects of nitrates mainly related to the control of the gut flora (Lundberg et al., 2004; Addiscott and Benjamin, 2004).

Accumulation of nitrates results from an imbalance between the uptake and translocation of nitrates by the xylem, and the reduction of these nitrates to ammonia which is subsequently rapidly incorporated into amino acids (Maynard et al., 1976). However, the internal nitrate concentration in the plant seems to be controlled by a self-regulatory mechanism exerted either by negative feedback control on the net nitrate uptake rate (Cardenas-Navarro et al., 1998) or by passive control on nitrate efflux (Scaife, 1989). The viewpoint that non-structural carbohydrates and nitrates have a complementary role in maintaining cell tugor (Blom-Zandstra and Lampe, 1985; Behr and Wiebe, 1988) offers a credible model for the plant nitrate regulation mechanism suggesting the accumulation of nitrates in the vacuole as an alternative osmoticum under low radiation

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conditions (Blom-Zandstra, 1989; Buwalda and Warmenhoven, 1999).

Accumulation of nitrates in lettuce has been shown to be affected by the soil texture and the source of fertilizer-N (Scaife et al., 1986; Gianquinto et al., 1992; Gunes et al., 1995), the NH_4-N-to-NO_3-N fertilizer-N ratio (McCall and Willumsen, 1998; Demsar and Osvald, 2003), the timing of fertilizer-N release (Tesi and Lenzi, 1998), the light intensity and duration (Behr and Wiebe, 1992; Chadjaa et al., 1999; Drews et al., 1995; Gaudreau et al., 1995), crop season (Gianquinto et al., 1992), and lettuce type and cultivar (Blom-Zandstra and Eenink, 1986; Siomos, 2000; Escobar-Gutierrez et al., 2002). In most types of lettuce, including the romaine type, the highest concentration of nitrates is normally observed in the external leaves (Corre and Breimer, 1979; Santamaria et al., 1999; Abu-Rayyan et al., 2004).

In countries of the Mediterranean basin, sheep manures are traditionally used as an organic fertilization source. Their recycling into soils low in organic matter, which are widespread in these regions, could benefit soil structure and long term fertility, and is also an alternative to inorganic fertilizers in the growing organic vegetable production business.

In this work we compare short and long term effects of organic *versus* inorganic sources of N on the growth and the accumulation of nitrates in the external leaves of a romaine lettuce. Composted sheep manure is used for organic fertilization whereas standard fertigation applied by farmers in the area is used for the comparison to inorganic fertilization. Fertilization from both sources is applied at a minimum, a medium and a maximum dose during three successive crop seasons in the same soil. Effects of crop season and long term cumulative fertilization on plant and soil are studied.

2. Materials and methods

2.1. Location and design of experiments

The experiments were carried out during three successive crop seasons: late spring (first, 6 April–17 May 2000), late autumn (second, 26 October–5 December 2000), and late-winter (third, 6 February–28 March 2001), at the N.AG.RE.F.-Olive and Horticultural Crops Institute of Kalamata, S.W. Peloponnese, Greece (37°03′N, 22°07′E, altitude 9 m). At the start of each crop season, a romaine lettuce crop cv. Corsica (Royal Sluis) was established in a polyethylene covered greenhouse (80–85% transparency to visible irradiation), with 60% side wall cover. The greenhouse soil was a sandy clay loam (SCL) with chemical characteristics presented in Table 1.

Seven long-term fertilization treatments were compared including: (i) three different organic fertilization treatments (*O-min*, *O-mid*, *O-max*) consisting of composted sheep manure (Table 1) applied at the start of each cultivation period, at doses of 1–3 l plant^{-1} respectively for the first and second crop seasons, and reduced by 33% for the third crop season (0.67, 1.34, and 2.01 l plant^{-1}, respectively), (ii) three different treatments of inorganic-N fertilization (*I-min*, *I-mid*, *I-max*) applied *via* drip irrigation during each crop season at doses corresponding to 87, 174 and 261 mg N plant^{-1} fertigation^{-1}, respectively (Table 2), and (iii) a control treatment in which no fertilizer was applied (*C*). The nutrient elements provided in total during each crop season by the organic and inorganic fertilization treatments are shown in Table 3. In all three inorganic fertilization treatments (I-min, I-mid, I-max) the soluble commercial fertilizers 15–30–15 and 20–20–20 plus micro-nutrients were applied, each at a rate of 0.25 g plant^{-1} fertigation^{-1}. Extra N fertilization was applied for the I-mid and I-max treatments with the inorganic fertilizer 34.5–0–0 (NH_4NO_3) at rates of 0.25 and 0.5 g plant^{-1} fertigation^{-1}, respectively.

Each experimental plot consisted of 54 lettuce plants placed in three rows (30 cm apart) of 18 plants (25 cm apart). The plants of the two outer rows and the three plants in each end of the middle row were kept as guard plants. There were five replicate plots per treatment and the experiment was set up as a randomized complete block design.

Table 1
Chemical characteristics of soil and sheep manure used in the experiments

	E.C. (mS cm^{-1})	pH	$CaCO_3$ (%)	O.M. (%)	C.E.C. (cmol$^{(+)}$ kg^{-1})	$N_{Kjeldahl}$ (g kg^{-1})	P_{Olsen} (mg kg^{-1})	K_{exch} (mg kg^{-1})
Soil	2.6	7.4	13.4	1.4	8.6	0.15	37.0	118.2
Manure[a]	21.7	8.0	8.4	72.0	36.0	1.64	1.4 (% d.w.)	1.8 (% d.w.)

[a] Manure dry weight = 201 g l^{-1}.

Table 2
Nutrient elements provided per fertigation by the inorganic fertilization treatments applied to lettuce

Fertilization treatment	Nutrient elements provided in each crop season							
	N (g plant^{-1} fertigation^{-1})	P (g plant^{-1} fertigation^{-1})	K (g plant^{-1} fertigation^{-1})	Mg (mg plant^{-1} fertigation^{-1})	Fe (mg plant^{-1} fertigation^{-1})	Mn (mg plant^{-1} fertigation^{-1})	Zn (mg plant^{-1} fertigation^{-1})	Cu
I-max	0.261	0.027	0.036	0	0.195	0.081	0.0163	0.0163
I-mid	0.174	0.027	0.036	0	0.195	0.081	0.0163	0.0163
I-min	0.087	0.027	0.036	0	0.195	0.081	0.0163	0.0163

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