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Mineral composition of durum wheat grain and pasta under increasing atmospheric CO₂ concentrations



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ABSTRACT

The concentrations of 10 minerals were investigated in the grain of 12 durum wheat genotypes grown under free air CO_2 enrichment conditions, and in four of their derived pasta samples, using inductively coupled plasma mass spectrometry. Compared to ambient CO_2 (400 ppm; AMB), under elevated CO_2 (570 ppm; ELE), the microelement and macro-element contents showed strong and significant decreases in the grain: Mn, -28.3%; Fe, -26.7%; Zn, -21.9%; Mg, -22.7%; Mo, -40.4%; K, -22.4%; and Ca, -19.5%. These variations defined the 12 genotypes as sensitive or non-sensitive to ELE. The pasta samples under AMB and ELE showed decreased mineral contents compared to the grain. Nevertheless, the contributions of the pasta to the recommended daily allowances remained relevant, also for the micro-elements under ELE conditions (range, from 18% of the recommended daily allowance for Zn, to 70% for Mn and Mo).

1. Introduction

The effects of increasing atmospheric CO_2 levels on the global climate are dramatically evident. As predicted by Easterling et al. (2007), the rise in atmospheric CO_2 is changing the global climate, with severe effects on mean temperatures and with altered patterns of global rainfall. According to the World Meteorological Organisation, the overall mean CO_2 concentration in the atmosphere in 2015 exceeded the symbolic limit of 400 parts per million (ppm) for the first time since measurements began (measured at Mauna Lowa Observatory, Hawaii). This was maintained through 2016, and was accompanied by a mean temperature increase of almost 1 $^{\circ}$ C (GHG-Bulletin, 2016).

To be able to predict the effects of these increasing atmospheric CO_2 concentrations on crop yields and quality, several studies have been conducted under conditions of free air CO_2 enrichment (FACE). Many studies have shown that the grain yield of C3 crops, such as wheat, increases under FACE (Badeck et al., 2013; DaMatta, Grandis, Arenque, & Buckeridge, 2010). Leakey et al. (2009) reviewed the earlier studies under FACE and calculated that with a CO_2 exposure of about 580 ppm, CO_3 plants (e.g., crops, trees) would increase their

photosynthetic carbon uptake by 19%-46%, even if their yield gain would be lower than expected. However, severe effects have been reported on the nitrogen and mineral concentrations in grain under these conditions: Fares et al. (2016) reported decreases of 7.0% and 13.3% for grain protein and gluten content, respectively; Myers et al. (2014) reported a similar decrease in grain protein content (6.3%), and also for Zn and Fe, with decreases of 9.3% and 5.1%, respectively. Fernando et al. (2012) instead reported greater decreases in both grain protein content, at around 12.7%, and grain Fe and Zn contents, at 10% and 22%, respectively. This might be of concern, as changes in grain composition are not only relevant for the nutritional value of cereal-based diets, but they might also affect the quality of the typical products that are obtained from the grain. This is the case in particular for durum wheat, where about 75% of the worldwide production (37 Mt; FAO, 2013) is used by the pasta industry. Similarly for the cous-cous and bread products that are intended for human consumption.

Recently, Fares et al. (2016) carried out FACE studies using CO_2 elevation to 570 ppm (ELE) for durum wheat, and they reported that the grain protein and gluten contents decreased (by 7.0%, 13.3%, respectively). Furthermore, the pasta quality worsened in terms of

Abbreviations: AMB, ambient CO₂ (400 ppm); ELE, elevated CO₂ (570 ppm); FACE, free air CO₂ enrichment; PCA, principal component analysis

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 Table 1

 Analysis of variance of the mineral content in the matching grain and pasta samples.

Preparation	Interaction	Significance									
		Na	Mg	P	K	Ca	Mn	Fe	Cu	Zn	Мо
Grain	E	< 0.0001	< 0.0001	n.s	< 0.0001	0.0183	< 0.0001	< 0.0001	n.s	0.0004	0.0339
	G	n.s.	n.s.	n.s.	n.s.	n.s.	0.0055	0.0002	n.s.	n.s.	n.s.
	$G \times E$	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	0.02	n.s.	n.s.	n.s.
Pasta	E	n.s.	n.s.	< 0.0001	0.0094	n.s.	0.0004	n.s.	0.0061	0.0369	0.0469
	G	0.0007	0.0162	< 0.0001	< 0.0001	n.s.	0.0008	< 0.0001	n.s.	0.0049	n.s.
	$G \times E$	0.046	< 0.0001	< 0.0001	0.0041	n.s.	0.0007	0.0046	n.s.	n.s.	n.s.

E, environment (ambient [AMB] or elevated CO_2 [ELE]); G, durum wheat genotype; n.s., not significant.

firmness under ELE (AMB, 406 g; ELE 373 g). These findings agree with other studies on wheat (*Triticum aestivum*). Fernando et al. (2015) established that the high-molecular-weight gluten protein sub-units in wheat grain decreased by up to 50%, and that this alteration in the grain protein quality was associated with lower bread quality. In an earlier study, the same authors reported an 11% decrease in the grain protein levels under ELE, which was associated with a bread volume decrease of 7% (Fernando et al., 2012). Similarly, studies conducted to date have shown that mineral concentrations can also be lower under ELE. Here, in C3 plants, Loladze (2014) showed that the overall mineral concentrations decreased by 8% (as the mean for 25 minerals), while Myers et al. (2014) reported mean decreases for Zn and Fe of 9.3% and 5.1%, respectively.

Accordingly, concerns have arisen for human nutrition, because dietary deficiencies of the basic micro-elements (e.g., Fe, Zn) represent a global public health problem (Myers et al., 2014). Above all, this indicates added risk for underdeveloped regions in countries such as India or Africa, where wheat and pulses are the main dietary sources for a large part of the rural population. Under such conditions, the risk of not reaching the minimum daily requirements for protein, Fe and Zn intake might have severe effects on public health (Myers et al., 2014). Furthermore, owing to an increase in people who prefer a diet based only on vegetables in the developed countries, this concern might also become a serious problem for public health. Thus, global declarations about "hidden hunger" and obesity are under widespread discussion in the current literature (Loladze, 2014; Myers et al., 2014), and are destined to become more of a global challenge.

The present study completes the description of the potential effects on the grain and pasta quality of durum wheat varieties caused by rising concentrations of CO_2 in the atmosphere. The durum wheat was thus grown under ELE conditions according to our previously published details (Fares et al., 2016). To the best of our knowledge, there have not been any studies of the effects of FACE on micro-element and macro-element levels in grain and pasta across a large number of durum wheat varieties, as in our studies here. As pasta represents a cheap food that is consumed worldwide, any changes in its nutritional profile need to be carefully evaluated to estimate the consequent variations in dietary mineral intake. To do this, we evaluated the effects of pasta processing on durum wheat grown under ELE, and evaluated the contributions to the recommended daily allowance (RDA) of the pasta samples, for each mineral element.

2. Materials and methods

2.1. Field materials

Ten durum wheat (*Triticum durum*) varieties with contrasting quality characteristics were selected for this study: Simeto, Ciccio, Claudio, Anco Marzio and Saragolla, as modern high-yielding varieties; Svevo and Aureo, as high-protein content varieties; and Cappelli, Creso and Ofanto, as varieties with a relevant role in the history of Italian durum wheat breeding. In addition, two lines selected from an

Ofanto × Cappelli recombinant inbred line population were included (RILl1, RIL28). The ELE conditions were installed on the experimental farm of the Genomics Research Centre of the Consiglio per la Ricerca in Agricoltura e l'Analisi dell'Economia Agraria (CREA) in Fiorenzuola d'Arda (44.927°N, 9.893°E). The soil was classified as silt-clay-loam (15% sand, 51% silt, 34% clay). Total nitrogen content was about 0.9%, organic matter was 1.8%, and K2O and P2O5 were 350 ppm and 12 ppm, respectively. The experimental units were plots of 2.2 m × 1.36 m. The ELE treatment (of 570 ppm CO₂) was applied to four octagons inscribed in circles of 14-m diameter. Each of these four ELE systems, and the four controls at ambient CO₂ (AMB 400 ppm), contained two replicates of the 12 genotypes. The ELE treatment was started on 16 November, 2011, and stopped when the leaves were senescent, on 14 June, 2012. The plots were fertilised with application of a N:P:K fertiliser at pre-seeding (45 kg for each N:P:K) and two top dressings with ammonium nitrate, of 52 kg N each. Treatments with herbicides and fungicides were applied according to local standard practices. The final harvest was on 2 July, 2012 (Fares et al., 2016).

2.2. Grain milling and pasta processing

The full description of the grain milling and pasta processing were published by Fares et al. (2016). In the present study, the grain of all of the genotypes previously described were analysed, while for the pasta samples, four were analysed accordingly: Saragolla and Simeto, as two varieties that were chosen from among those that did not shown significant differences in mineral compositions across these two environments; and Cappelli and Ciccio, which instead significantly differed for the same minerals. Moreover, we evaluated the mineral content of 100 g of pasta serving derived from the grain produced under AMB and ELE, while noting the reference values for each mineral (i.e., RDA) (EEC, 2008).

2.3. Determination of macro-elements and micro-elements

For the determination of the contents of the mineral macro-elements Na, K, P, Ca and Mg and micro-elements Mn, Fe, Cu, Zn and Mo of both the grain and the pasta, the dried samples were milled (Pulverisette 7; Planetary Micro Mill, Classic Line, Fritsch) using an agate jar and balls. Then 20 mg of each sample was used for the analysis (as four replicates for the grain samples, and three replicates for the pasta samples).

The macro-elements and micro-elements were determined on samples digested and diluted to 50 mL with high purity deionised water, in polypropylene disposable tubes, as reported by Ficco et al. (2009). Then, their content was analysed using inductively coupled plasma mass spectrometry (Agilent 7700x; Agilent Technologies, Italy), equipped with an auto-sampler (ASX-500), as described by Hansen et al. (2009), with minor modifications. The inductively coupled plasma mass spectrometry was tuned to standard mode and with collision gas (He) to remove many of the simple solvents and the argon-based polyatomic spectral interference. The plasma power was operated at 1550 \pm 50 W, and the carrier and make-up gases were typically set at

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