Gene expression and nutritional analyses revealed the positive effects of EXPANDO® on ripening, yield and fruit quality of tomato plants (*Solanum lycopersicum*)

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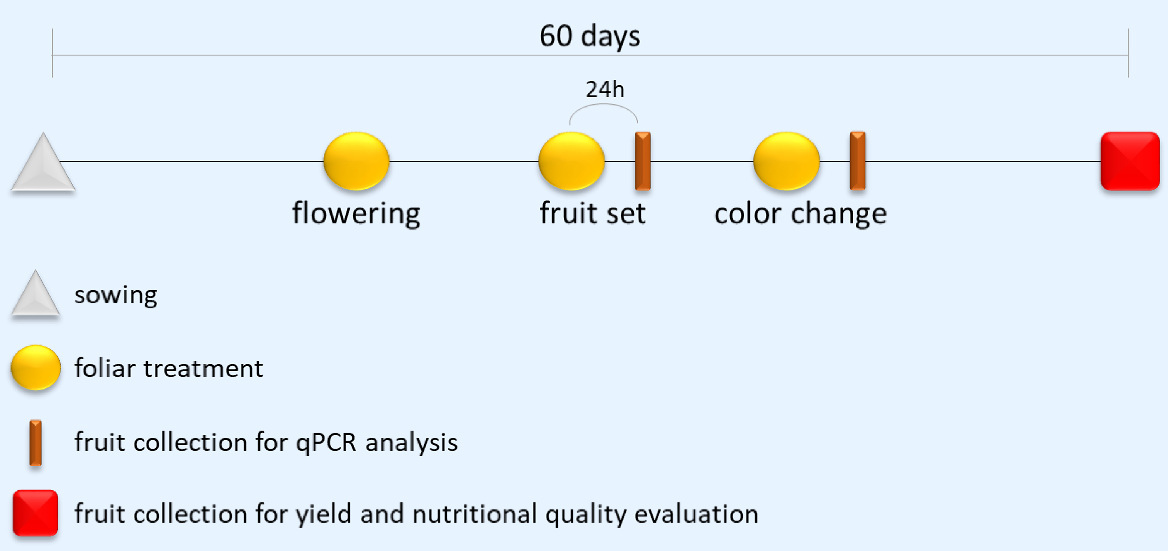
INTRODUCTION AND AIM OF THE WORK

Tomato (*Solanum lycopersicum*) is one of the most important annual crop worldwide. Due to its large demand, the increase of yield and fruit quality parameters represent an important focus for scientific research (1). The world population is growing fast and, moreover, the consumers tend to prefer foods cultivated in an eco-sustainable way because they are perceived to be safe, healthy and environmentally friendly (2). Biostimulants could satisfy both these needs, thanks to the absence of harmful substances, the low application doses and their ability to increase crop yield and quality (3, 4).

In this study, tomato plants were treated with EXPANDO®, a biostimulant based on mineral components, seaweed and yeast extracts, with the aim to evaluate its potential influence on fruit ripening, yield and nutritional quality.

MATERIALS AND METHODS



qPCR for expression analysis of genes involved in fruit ripening and quality

Digital scale and **caliber** for yield and fruit size evaluation

GC-MS for unsatured fatty acid analysis

Microwave plasma atomic-emission spectrometer for mineral content analysis

RESULTS

1 qPCR analysis

Forty-eight genes involved in fruit ripening and quality were analyzed at fruit-set and color change phase. Tomato plants treated with EXPANDO® showed a significant up-regulation of 16 genes related to expansins, chlorophyll and carotenoid biosynthesis a both phases. The most significant up-regulated genes are reported (Fold Change from 1.2 to 4, p<0.05) in Table 1

Table 1 Main up-regulated genes in fruits treated with EXPANDO® at fruit-set and colour-change. These are grouped in Expansin-related genes, Chlorophylls biosynthesis-related genes and Carotenoids biosynthesis-related genes. The Fold Change (FC) obtained from qPCR at both phases are reported. Gene ID is from www.ncbi.nlm.nih.gov.

GENE	GENE ID	FUNCTION	FC fruit-set	FC color-change	
Exp2	543582	Fruit-size improvement	1.7	1.6	Expansin-related genes
Exp5	543558		1.4	3	
Exp6	36276		1.2	1.3	
Golden 2-like protein	101055613	Transcription factor involved in the regulation of chloroplast and chlorophyll production levels	1.2	1.2	Chlorophylls biosynthesis-related gened
Protochlorophyllide oxidoreductase	101244717	Involved in the conversion of protochlorophyllide to chlorophyllide, a key step in the chlorophyll biosynthesis	3.4	1.7	
chlorophyll binding protein	101266182	Part of the light-harvesting complex of Photosystem II	1.2	4	Carotenoids biosynthesis-related genes
15-cis-phytoene desaturase	101244544	Biosynthesis of lycopene and ζ-carotene	1.4	1.5	
zeta carotene desaturase	543629		1.2	1.2	

2 Yield and fruit-size

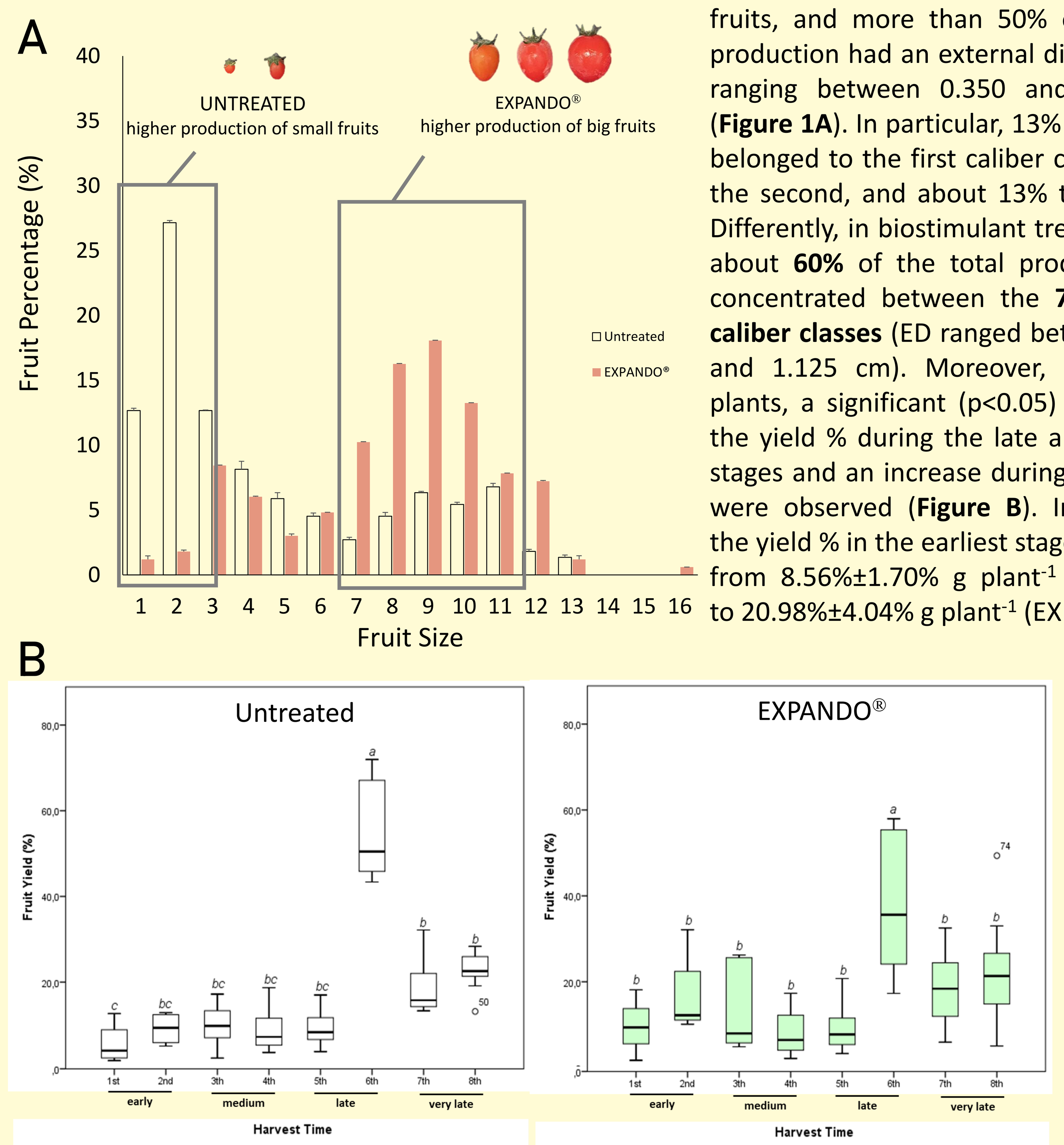


Figure 1. A: Size of the fruits produced by untreated tomato plants, or treated with EXPANDO®. The x-axis reports the caliber classes in which the fruits were grouped according to their external diameter. **B:** Fruit percentage (%) produced by tomato plants during the harvesting time (30 days). After the appearance of the first ripe fruits, the fully red-colored fruits were harvested every 4 days. Within the same panel, the different lowercase letters on the top of each boxplot indicate significant differences at p<0.05, as measured by Tukey's multiple range test. The letter "a" denotes the highest value.

Untreated plants produced very small fruits, and more than 50% of the total production had an external diameter (ED) ranging between 0.350 and 0.600 cm (**Figure 1A**). In particular, 13% of the fruits belonged to the first caliber class, 27% to the second, and about 13% to the third. Differently, in biostimulant treated plants, about **60%** of the total production was concentrated between the **7th and 10th caliber classes** (ED ranged between 0.825 and 1.125 cm). Moreover, in treated plants, a significant (p<0.05) decrease in the yield % during the late and very late stages and an increase during early times were observed (**Figure B**). In particular, the yield % in the earliest stages increased from 8.56%±1.70% g plant^{-1} (Untreated) to 20.98%±4.04% g plant^{-1} (EXPANDO®).

3 Unsatured fatty acid analysis

Table 2. Fatty acid % composition. Within the same row, different lowercase letters indicate statistical differences among the samples, as measured by *t*-test (p<0.05).

Fatty acid % composition	Untreted	EXPANDO®
C16:3ω3	0.202±0.026 b	0.866±0.009 a
C16:1ω7	0.431±0.044 b	0.655±0.102 a
C16:1ω10	0.089±0.017 a	0.155±0.012 a
C16:0	19.734±0.935 a	19.604±0.150 a
C18:2ω6	48.984±2.988 a	37.964±0.013 b
C18:1ω9	1.410±0.122 b	2.352±0.039 a
C18:0	4.128±0.181 a	4.060±0.045 a

The main fatty acid detected was linoleic acid (C18:2ω6), followed by oleic acid (C18:1ω9) and palmitic acid (C16:0). After the treatment with the biostimulant, strong changes in the % content were observed. In particular, the % content of roughanic acid (C16:3ω3), palmitoleic acid (C16:1ω7), oleic acid (C18:1ω9), elaidic acid (C18:1ω9), and sapienic acid (C16:1ω10) increased in parallel to the decrease of linoleic acid (C18:2ω6) (Table 2).

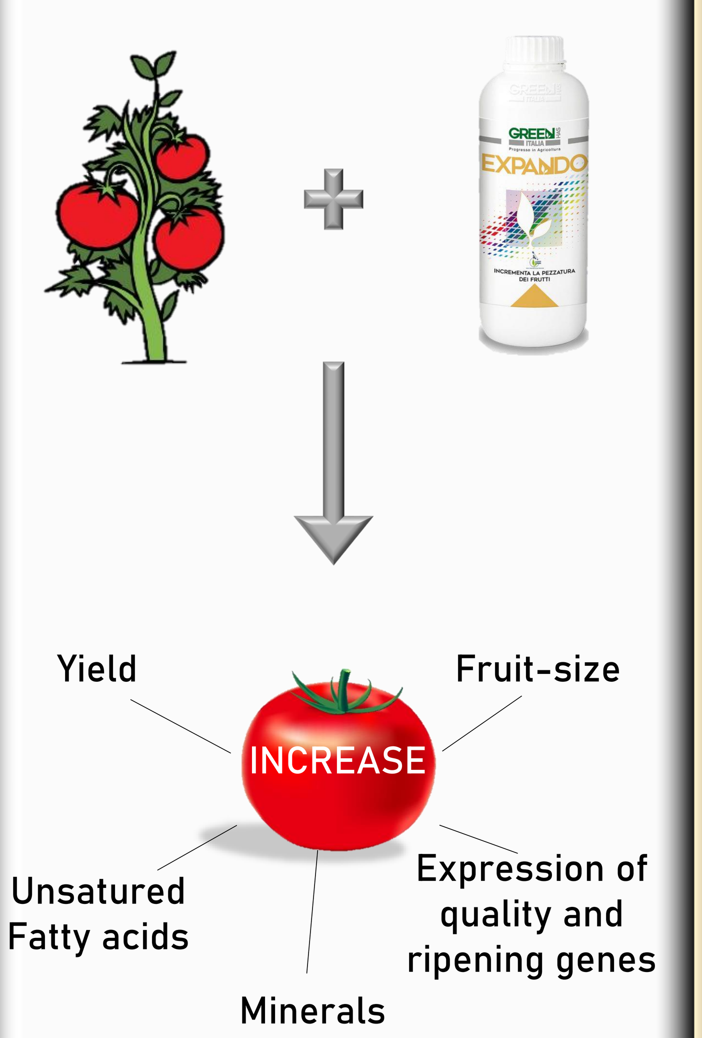
4 Mineral content

Table 3. Mineral content. Within the same row, different lowercase letters indicate statistical differences among the samples, as measured by *t*-test (p<0.05).

Mineral content (*mg g^{-1} FW*)	Untreated	EXPANDO®
K	541.671±0.772 b	549.724±4.953 a
Na	10.874±0.283 b	13.535±0.471 a
Ca	27.653±0.469 a	23.612±0.425 b
Mg	20.574±0.053 a	21.027±0.375 a
P	113.791±2.546 b	126.579±2.487 a
Cl	2.753±0.033 a	2.394±0.080 b
Fe	0.505±0.022 b	0.577±0.014 a
Cu	0.096±0.003 b	0.117±0.008 a
Zn	0.089±0.005 b	0.111±0.006 a
Mn	0.145±0.002 b	0.164±0.004 a
Si	0.506±0,013 b	0.625±0.021 a
B	0.046±0.005 a	0.052±0.003 a
Mo	0.013±0.001 b	0.016±0.002 a

In our experimental conditions, independently from the treatments, K was the most abundant in all the analyzed fruits, ranging between 73 and 75% of the total mineral content; meanwhile, Na was only about 1.5%. When tomato plants were treated with the biostimulant, MP-AES analysis revealed changes in mineral profile. In particular, the fruits harvested from biostimulant-treated plants were enriched in K, Na, P, Fe, Cu, Zn, Mn, and Mo that showed statistical differences compared to the control (Table 3).

CONCLUSIONS



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