TITLE: Effective Microorganisms (EM) in hydroponic lettuce (*Lactuca sativa* L.) intended for healthy functional bioactive enrichment.

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ABSTRACT:

Effective Microorganisms (EM) is a natural biological fertilizer and a promising alternative with distinct and complementary ecological function, which can reduce the nitrogen fertilizers, minimizing the accumulation of carcinogenic nitrate in edible plant tissue. The study evaluated the growth and nitrate accumulation in hydroponic lettuce (*Lactuca sativa* L.) combining EM treatment (0.1 and 1% of commercial EM-1®) under reduced nitrogen source (10 and 25% of recommended nitrogen value, offered as urea solution) by Nutrient Film Technique (NFT). Summer and autumn season data - macro and micronutrients in plant tissue, growth parameters, microbial counting and NGS (*Next Generation Sequencing*) in root were analyzed. The nitrate content in tissue was reduced in a factor of 90% in both seasons, with lower EM dose (0.1%) supplemented with 25% of nitrogen as urea form showing better performance. Summer culture showed lower nitrogen content than autumn, and phosphorus, zinc and iron were significantly higher in EM-treated plants ($p \leq 0.05$). Summer culture showed better shoot growth, although of stress damage due to long and high temperature period. Remarkable effectiveness under such extreme temperature could be attributed to EM action, and an increase of 156% in shoot growth was observed when compared with control, suggesting advantageous adaptation provided by EM association ($p \leq 0.05$). Microbial analysis showed good root adhesion of microorganisms composing EM (10^5 and 10^6 CFU/g for yeast and lactic acid bacteria, respectively), with abundant mesophilic and psychotropic microorganisms in EM treated plants ($p \leq 0.05$). The molecular sequencing showed a predominance of colonization by EM in the root portion, a fact that would provide competition with pathogen, combined with the production of bioactive and phytohormones in symbiosis in the rhizosphere, favoring plant growth. Summarizing, the combined use of EM and urea reduced nitrate content in plant tissue, and its microbial interaction improved growth and adaptation in stressed situation. In addition, such system could be a vehicle for food enrichment using bioactive through natural administration arrangement, i.e., replacing the artificial system consisting of micro or nano-capsules by low-cost plant cell system which can efficiently introduced in routine popular nourishment.

Keywords: effective microorganisms, nitrate, hydroponic, next generation sequencing

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