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Evaluation of the phytostimulant activity of microbial biomass containing *Bacillus subtilis*

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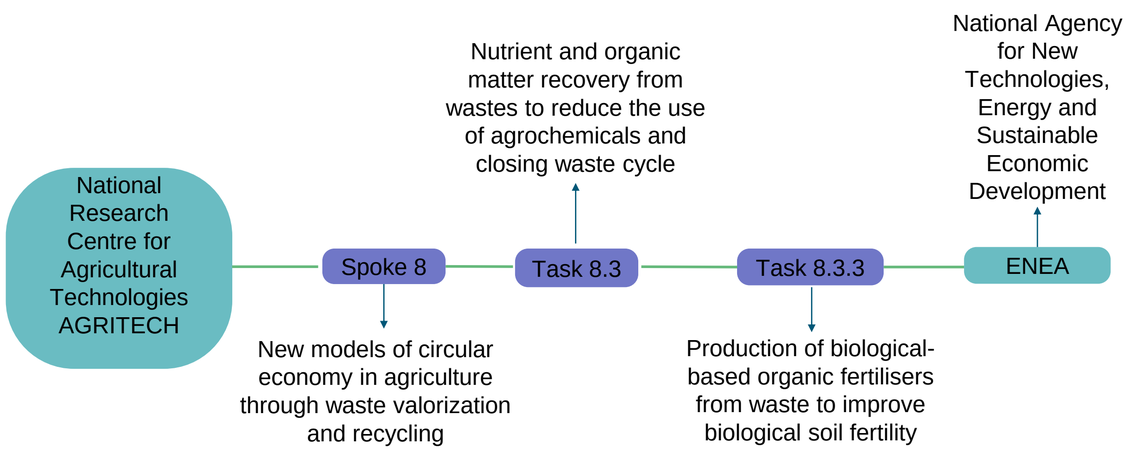
SPOKE and TASK



ABSTRACT

The research of more sustainable agricultural practices to improve plant productivity while reducing chemical pesticides and synthetic fertilizers use is one of the greatest challenges for the future agriculture.

Plant growth promoting microorganisms (PGPMs) are an interesting biotechnological tool. They can produce different positive effects on plants health. PGPMs makes the host plant less susceptible to disease, stimulate the plant nutritional processes while improving nutrient use efficiency, tolerance to abiotic stress, qualitative characteristics, and nutrients availability (Elnahal et al., 2022; Wei et al., 2020).

Bacillus subtilis [(Ehrenberg) Cohn] is a Gram-positive, non-pathogenic, ubiquitous bacterium known in agriculture as a biocontrol agent (Trupo et al., 2023) and a member of the Plant Growth Promoting Rhizobacteria (PGPR) (Franco – Sierra et al., 2020; Mahapatra et al., 2022) .

Respecting the principles of the circular economy, this research aimed to produce a microbial biomass containing *B. subtilis* ET–1 using agro – industrial waste as a breeding substrate. The phytostimulant attitude of the bacterium was tested *in vivo* on Basil (*Ocimum basilicum* L.) plants grown in a ventilating lamp system.

Results shown that *B. subtilis* ET -1 can produce positive effects on growth and yield of the treated plants.

MATERIALS and METHODS

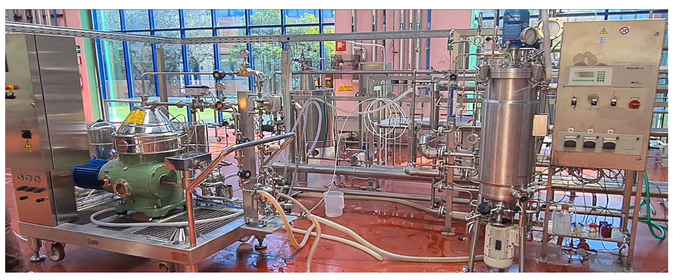
The experimentation consisted of two phases:

1) Microbial biomass production process

- *B. subtilis* ET-1 was grown in a 50-L stirred-tank bioreactor (B. Braun Biotech International, Germany) with 35 L of a designed substrate composed of agro-industrial waste.
- At the end of the culture, bacterial cells were separated from broth medium by a Seital centrifuge unit (SPX Flow Inc.).
- To develop a stable formulation of *B. subtilis* strain ET-1, the cells with the addition of 10 % (w/v) skimmed milk powder were spray-dried by APT-2.0 Spray Dryer (Yánez-Mendizábal et al., 2012).

2) Evaluation of the microbial biomass phytostimulant activity on Basil plants

- Basil plants were farmed under a ventilating lamp system.
- Three thesis were considered: plants treated with water, plants treated with ammonium sulphate and plants treated with microbial biomass.
- According to the thesis, Basil plants were treated with a total of 0,600 g of microbial biomass or ammonium sulphate through irrigation.
- To evaluate the phytostimulant effect, phenotypic observations, height, number of knots, fresh weights, dry weights and Chlorophyll Content Index (C.C.I.) of the three thesis were compared.


Centrifugal separator and 50 L fermenter for growth of *B. subtilis* ET– 1


Centrifuged


Spray - dryer


Dry biomass containing *B. subtilis* ET-1

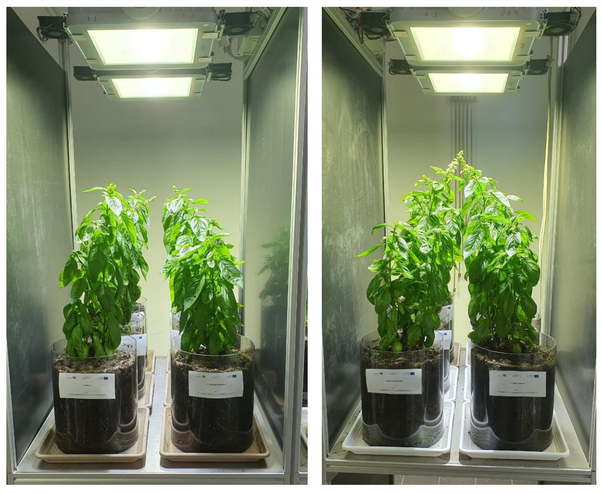

Evaluation of phytostimulant activity on Basil grown under ventilated lamp


C.C.I. and biometric determinations

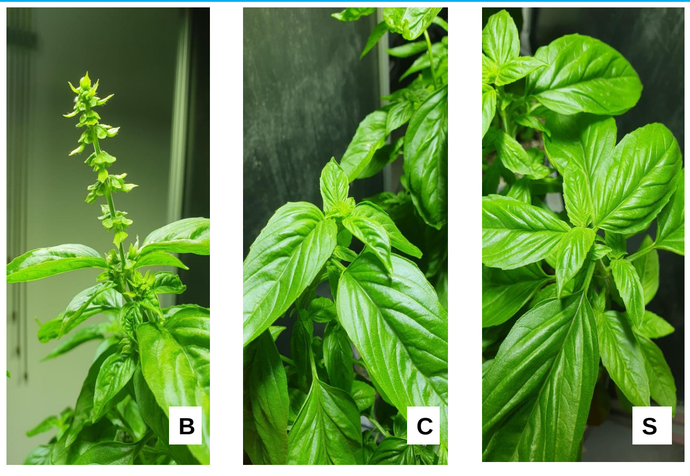
RESULTS

In this research:

- Dry biomass yield was 25 g /L of centrifuged culture broth.
- The obtained microbial biomass had a vitality of 7.2 × 10^8 CFU/g post spray-drying.
- Microbial biomass did not caused stress on plants.
- The plants treated with *B. subtilis* ET-1 were more vigorous and gave better results in terms of development and branching of the root system and speed of emission of the floral scape compared to the other two theses.


Basil plants at the end of the crop cycle


Detail of the development and branching of the root system of Basil treated with microbial biomass.



Differences in the rate of flower scape emission in plants treated with microbial biomass (B), water (C) and ammonium sulfate (S).

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