Evaluation of the Soil Fertilization with Encapsulates of *Chlorella vulgaris* and a *C. vulgaris/Pseudomonas putida* Consortium to the Growth of Meadow Clover Plantlets

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INTRODUCTION

Chlorella vulgaris is a chlorophyte, unicellular microalga, often used because it is very rich in carbohydrates and proteins, and some strains also produce growth factors (Ördög *et al.*, 2004). Besides, organisms can be very useful as soil conditioners, improving soil stability and capacity to retain water (Mettig *et al.*, 1990). Moreover, if microorganisms are encapsulated, they are protected from several environmental stress types; furthermore, they can gradually release nutrients to the soil (Bashan *el al.*, 2002)

In this work, a consortium with *C. vulgaris and different strains of bacteria* were tested, including formulations encapsulated in maltodextrin (MD), gelatine (G) and Arabic gum (GA).

Meadow clover dried plantlets

METHOLOGIES

Organisms used
- T3 – *Pseudomonas putida*
- E1 – *Serratia proteamaculans*
- *Chlorella vulgaris*

Plant growth conditions
- light: 54.73µEs^{-1}m^{-2}, constant
- temperature: 25ºC, constant
- watered every other day

Biomass treatment
- centrifugation: 9000 rpm, 10 min, 4-10ºC
- freeze-drying
- incorporation in encapsulates
  - maltodextrin, gelatine, (with or without) freeze-dried biomass
  - maltodextrin, Arabic gum, (with or without) freeze-dried biomass
  - Biomass: one week grown cultures of *C. vulgaris* alone or with bacteria

Obtaining encapsulates by spray drying
- Inlet temperature 150ºC
- Outlet temperature ~90ºC
- Aspirator 80-85%
- Peristalsis pump 24-26%

Composition of soils for growing meadow clover plantlets
- 70% sand : 30% turf : 1% $CaCO_3$: 0.06% dried (encapsulated) biomass
- 20 seeds per box, in triplicates
- 1st trial - *C. vulgaris* and *P. putida*, simply dried or encapsulates
  2nd trial - *C. vulgaris* and *S. proteamaculans*, simply dried or encapsulates

After seed germination, percentage of seed germination was registered; plantlets grew for a two-week period, and then root system and stem was measured (cm) and weighed (mg).

Composition of soils for growing meadow clover plantlets
- Anova/Manova from Statistica 6.0 Program (StatSoft Inc.)
- Minimum significant difference (Fisher method) was used to separate averages, $p<0.05$

RESULTS AND DISCUSSION

When working with the consortium *Chlorella/Pseudomonas* (T3), and when biomass is compared, three of the fertilizers provided the best conditions for plants to grow (fig 1), MD/GA/A 1:1:1 and MD/GA/A 1:2:1, and also MD/G/AB 1:2:1.

Results are somewhat different when dealing with *Chlorella/Serratia*: addition of algae to the encapsulates enhanced growth, but stem was higher when dried algal biomass was used, without being encapsulated.

Also, best proportions were obtained with MD/GA/A 1:2:1 and MD/G/A 1:2:1 (fig 1A). Nevertheless, dried biomass of roots and stems/leaves showed to be a more reliable parameter – a longest root or stem does not mean a greater absorption area, either for water and mineral salts, or for light.

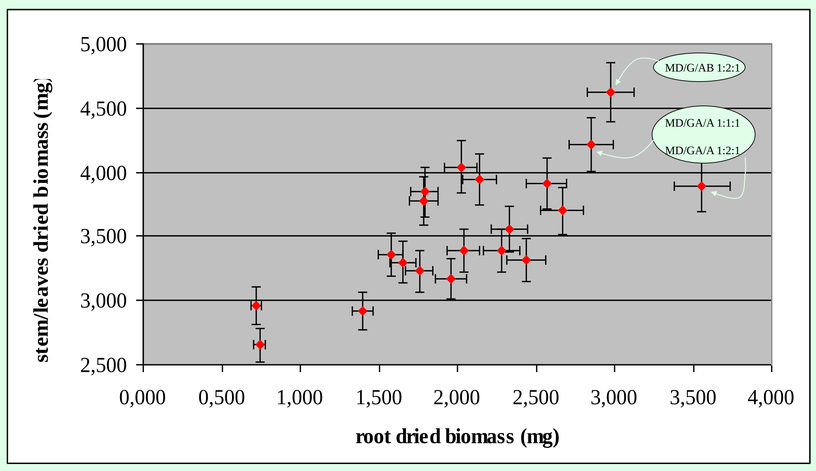

Figure 1. Graphic represents average dried biomass of plantlets grown with the consortium *Chlorella/Pseudomonas*.

With the consortium *Chlorella/Serratia* and looking to the figure 2B one can observe that plants with best proportions in the biomass stem/leaves-root system were obtained when included in the encapsulates MD/**G**/AB 1:**2**:1 and MD/GA/AB 1:1:1, and also with a soil fertilized with spray dried powder of *Chlorella+Serratia* (E1).

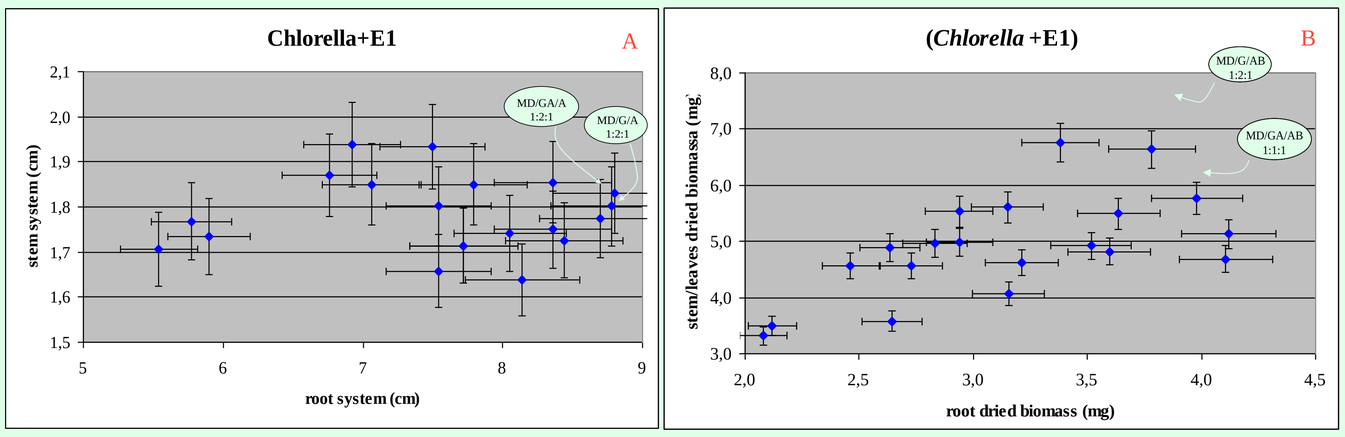

Figure 1. Graphic A represents length of roots and stems, and graphic B average of dried biomass of plants grown with the consortium *Chlorella/ Serratia*.

Comparing the results of *Chlorella/Pseudomonas* (T3) and *Chlorella/Serratia* (fig 3), one can observe that the second consortium was the one which was best for the development of meadow clover plantlets, specially when included in MD/GA/AB 1:1:1 encapsulates (Chl+E1), Again, and despite the good growth in MD/G/AB 1:2:1, with (Chl+T3) *Chlorella/Pseudomonas*, most developed plants were obtained with *Chlorella/Serratia* (Chl+E1) as MD/G/AB 1:2:1.

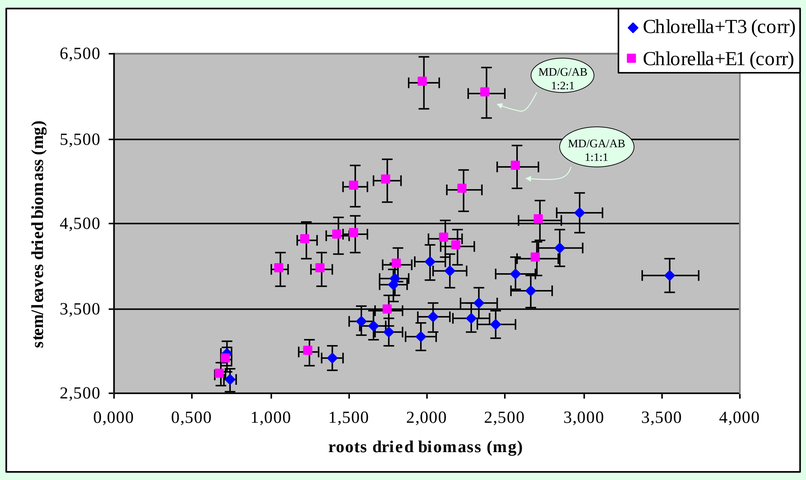

Figure 3. Results in the graphic serve to compare development of plants induced by the consortia *Chlorella/Pseudomonas* (T3) and *Chlorella/ Serratia* (E1).

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