Growth and Micropropagation Assays of *Salicornia* and *Sarcocornia* Plants

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INTRODUCTION

Salicornia and *Sarcocornia* are two succulent, halophytic plants used in salads or as pickles, hence the name "pickleweed" . Their seeds are very rich in edible oils, highly polyunsaturated, linoleic acid contributing with 70% to the fatty acids content. Salicornias also have high amounts of iodine, phosphorus, zinc, and vitamins A, C and D.

Due to the increasing interest in the food industry and also because it is urgent to replace these plants in their habitats, it is also natural that one thinks in their quick and efficient propagation.

At a laboratorial level, micropropagation is the process used to obtain plants with the same phenotypic and genetic characteristics, usually involving manipulation of the culture media, adding (or not) some growth regulators, to allow development of stems or roots of the cultivated explants.

EXPLANTS → organized structures (meristems, apex, buds)

→ differentiated cells (pieces of leaves, stems or roots)

+ auxins (IAA, 2,4-D) → induction of differentiation → new organization

→ root production

→ stems production + cytokinins (2iP, kinetin)

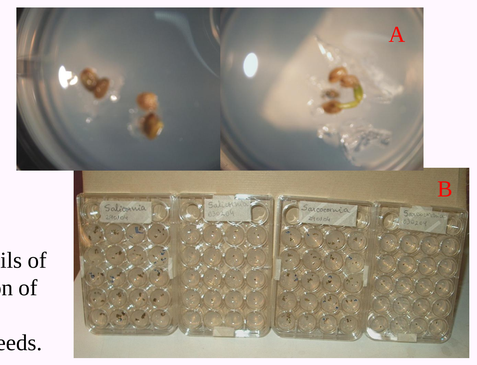

Figure 1. Details of the germination of *Salicornia*/ *Sarcocornia* seeds.

METHODOLOGIES

Seeds disinfection

- ethyl alcohol 70%
- sterilised deionised water
- sodium hypochlorite 2.5% + 0.5% tween 20
- sterilised deionised water, 3x
- benomil solution 1%

Figure 2. Another aspect of the germination: when adding the growth regulators.

Germination

- in Petri dishes, with H&A medium (fig 1A) or in tubes (fig 1B),
- embedded (or not) in growth regulators GA3, 100mg l^{-1}, or salicylic acid, 7mg l^{-1} (fig 2),
- put to germinate in media without hormones or with 0.1mg l^{-1} IAA + 0.5mg l^{-1} BAP (Mei *et al.*, 1997), or 2mg l^{-1} NAA + 1mg l^{-1} BAP + 1 GA3 (Libik *et al.*, 2005)

Figure 3. Picture represents plantlets growing in tubes and in glass flasks, in H&A medium

Growth of plantlets

- in tubes (fig 3A) or in small glass flasks (fig 3B)
- in (2)H&A medium (for a better growth)
- growth chamber, under constant temperature (25ºC) and illumination (29.18 $\mu Es^{-1}m^{-2}$)

Induction of organogenesis

Stem	1st assay	IBA, NAA, 2,4-D	0.025mg l^{-1}
		kin	0.5, 1.0, 1.5, 2.0 mg l^{-1} (fig 4A)
	2nd assay	IAA, IBA, 2,4-D	0.5, 1.0 mg l^{-1}
		kin	0.5, 1.0, 1.5, 2.0 mg l^{-1} (fig 4B)
Root	1st assay	IBA, 2,4-D	0.625 mg l^{-1}
		kin	0.037 mg l^{-1} (fig 5A)
	2nd assay	IBA	1.825 mg l^{-1} (fig 5B)

AGRADECIMENTOS – Projecto Agro, Produção Experimental da Planta Halófila *Salicornia*. UE/Min. Agricultura.

RESULTS AND DISCUSSION

Due to the poor results obtained in the first micropropagation attempts, probably because of the stress induced by the salinity (2% NaCl, w/v) or because explants did not have the capacity to obtain nutrients directly from the culture medium, the concentration of the nutrients was doubled, and, for the experiments with explants of *Salicornia/Sarcocornia*, medium was supplemented with casein hydrolysate (Prehn *et al.*, 2003; Ahmad & Anis, 2005).

Figure 4. Organogenesis assays with *Salicornia* explants to induce stem growth.

Figure 5. Organogenesis assays with *Salicornia* to induce root growth.

When nitrogen and phosphorus were increased plant answer was positive and plantlets begin to show a better growth (fig 6A and B).

Figure 6. Salicornias show better growth when in doubled concentration of nitrogen and phosphorus.

Explants are also responding well to the addition of casein hydrolysate (HC) (100 and 200 mg l^{-1}), to which BAP (benzyl aminopurine) was added, best growth obtained with 200 mg l^{-1} HC plus 1 mg l^{-1} BAP.
A good answer was also obtained when using explants with 2-3 segments, either in a supplemented medium or not (fig 7).

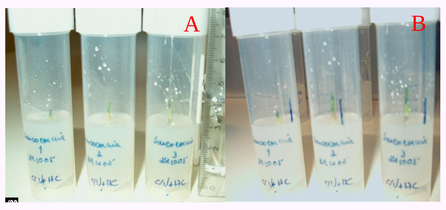

Figure 7. Explants of *Salicornia* with 1-3 segments, one week (A) and two weeks (B) after being cultivated in 2H&A medium with HC and vitamins.

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