



TISSUE CULTURE INITIATION AND PLANT REGENERATION IN *HEUCHERA* SP.

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Abstract: *Heuchera* sp. is an evergreen plant with great ornamental potential due to the diversity of colour and morphology of the leaves and the occurrence of inflorescences. The aim of this study is to establish the influence of plant species and explant on the regenerative potential of *Heuchera* sp. at the initiation of *in vitro* culture. The starting material used for the initiation of culture consists of petiole and leaf explants and shoot buds by two genotypes of *Heuchera*: *H. villosa* 'Silver Gumdrops' and *H. x hybrida* 'Timeless Orange'. In the asepsis stage, two solutions for the asepsis of shoot explants, 15% and 20% sodium hypochlorite, were tested for 20 min. Culture initiation was carried out on Murashige & Skoog (1964) culture medium, standard or half-formulated and with 1 mg/l BAP. The regeneration rate from shoots was 77.78% for 'Silver Gumdrops' and 63.33% for 'Timeless Orange'. Petiole explants resulted in 97.78% callusogenesis for 'Silver Gumdrops' and 85.56% for 'Timeless Orange'. Micropropagation proves to be the most efficient way to propagate this genus, due to the fact that seed propagation is poor and vegetative propagation offers low multiplication rates in long time intervals.

Introduction

The genus *Heuchera* comprises more than 53 species of herbaceous perennials native to North America (Wells, 1984). This plant is cold-resistant (-35° Celsius) and is recommended for cultivation in colder areas where it can overwinter in the field (Drăghia et al, 2014). *Heuchera* can be propagated by seed, bush splitting and tissue culture (Xu et al, 2022). Previous studies on micropropagation of the genus *Heuchera* have reported good results when starting *in vitro* cultures from shoot tips and petiole (Zhao et al, 2017). The culture medium was supplemented with cytokinins (BA or KIN) (Stapfer & Heuser, 1986) or with a hormonal balance including auxins (NAA and BA) (Miura et al, 1995).

Material and method

The biological material for the initiation of tissue cultures is represented by young plants, about 6 months old, with a healthy appearance, without signs of disease or pests. Explants were leaf, petiole and shoot fragments from two *Heuchera* genotypes:

H. villosa 'Silver Gumdrops' and *H. x hybrida* 'Timeless Orange'.

For the **asepsis** of the explants, in order to determine the optimal concentration of sodium hypochlorite (15% or 20%) for each type of explant (petiole, leaf and buds) and genotype, the asepsis treatment was organized on the model of a 2 x 3 x 2 three-factor experiment.

The study of the behavior of explants in the **initiation** of *in vitro* culture was organized on a 2 x 3 x 3 three-factor experiment, analysing the interaction between genotype, explant type and culture medium (½ MS = Murashige & Skoog (1962) with half concentration; MS = Murashige & Skoog (1962); MS1 = MS + 1 mg/l BAP) on organogenesis process.

Conclusions

Both *Heuchera* genotypes are suitable for *in vitro* mass propagation. The *Heuchera* stem buds are the prolific explants for culture initiation. The composition of the culture medium did not show a defining influence on plantlets regeneration, although the 1/2 MS medium without growth regulators proved to obtain the lowest regeneration rates and shortest plantlets compared to MS or MS1 where plantlets were significantly more vigorous.

Results and discussions

The effectiveness of asepsis treatment (Figure 1) was recorded 2-7 days after the inoculation of explants. The biometric data on the organogenesis process (Figure 2-4) were taken 60 days after culture initiation.

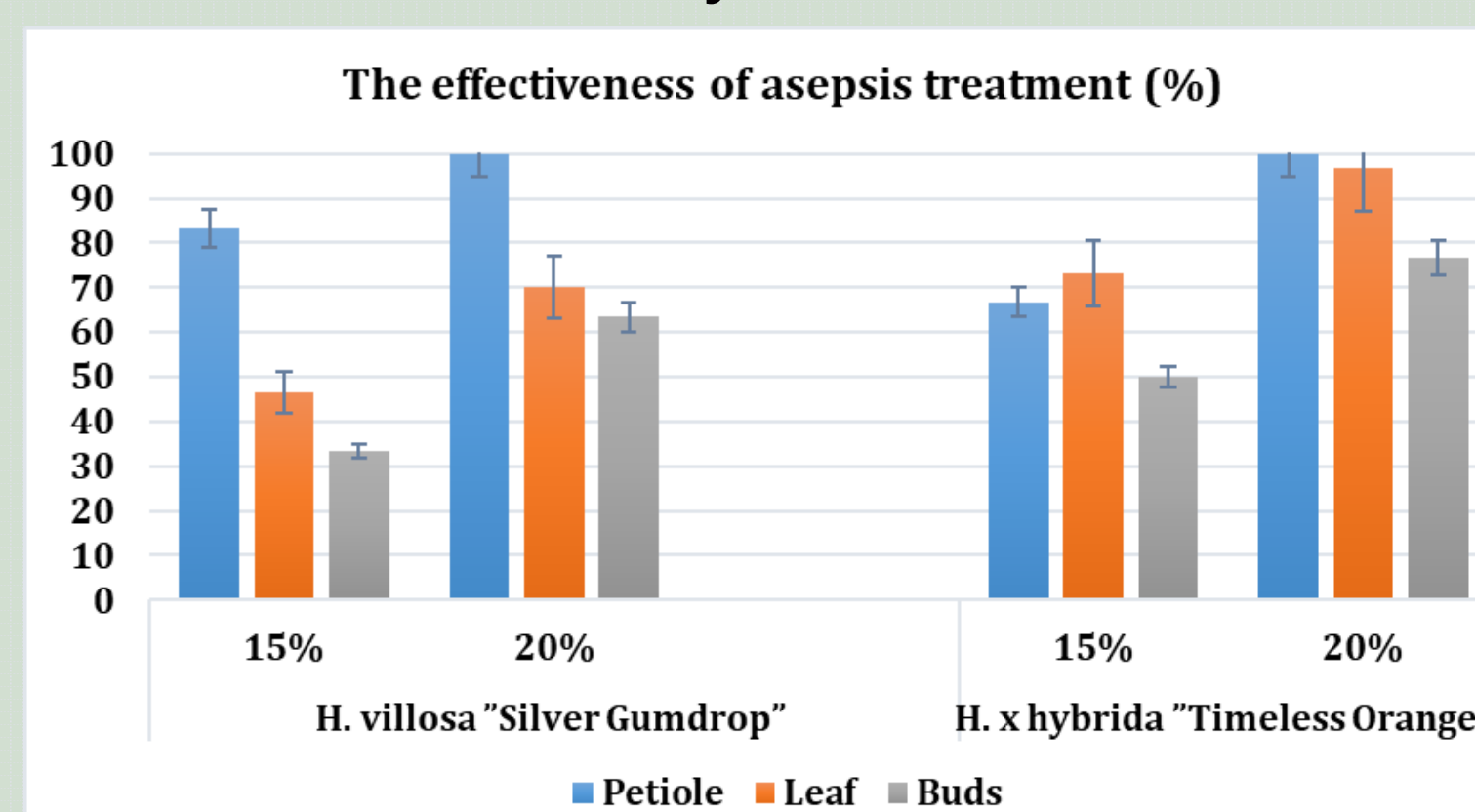


Figure 1. The interaction of genotype, explant type and sodium hypochlorite concentration of effectiveness of asepsis treatments.

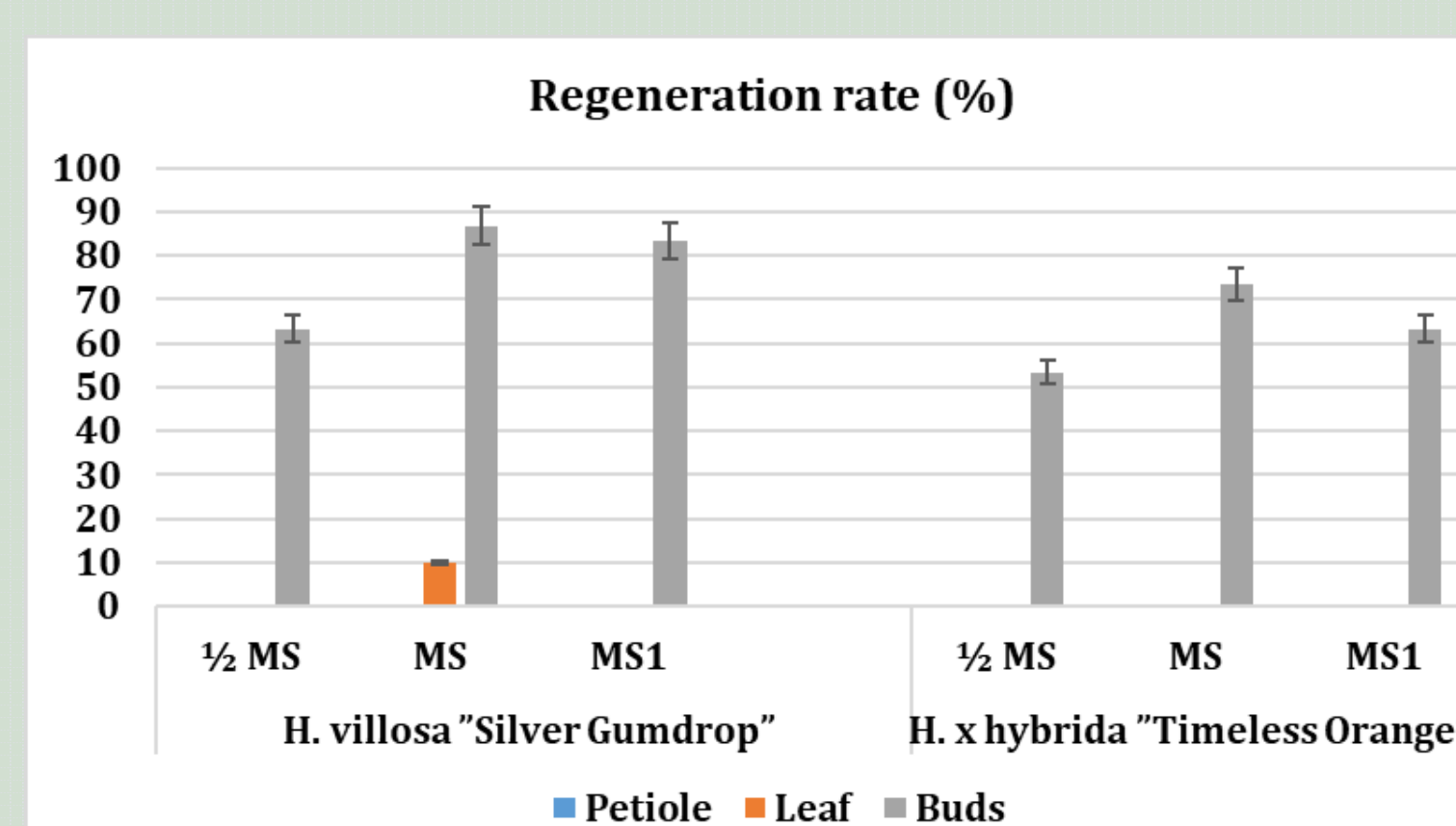


Figure 2. The interaction of genotype, explant type and culture media on regeneration rate (%).

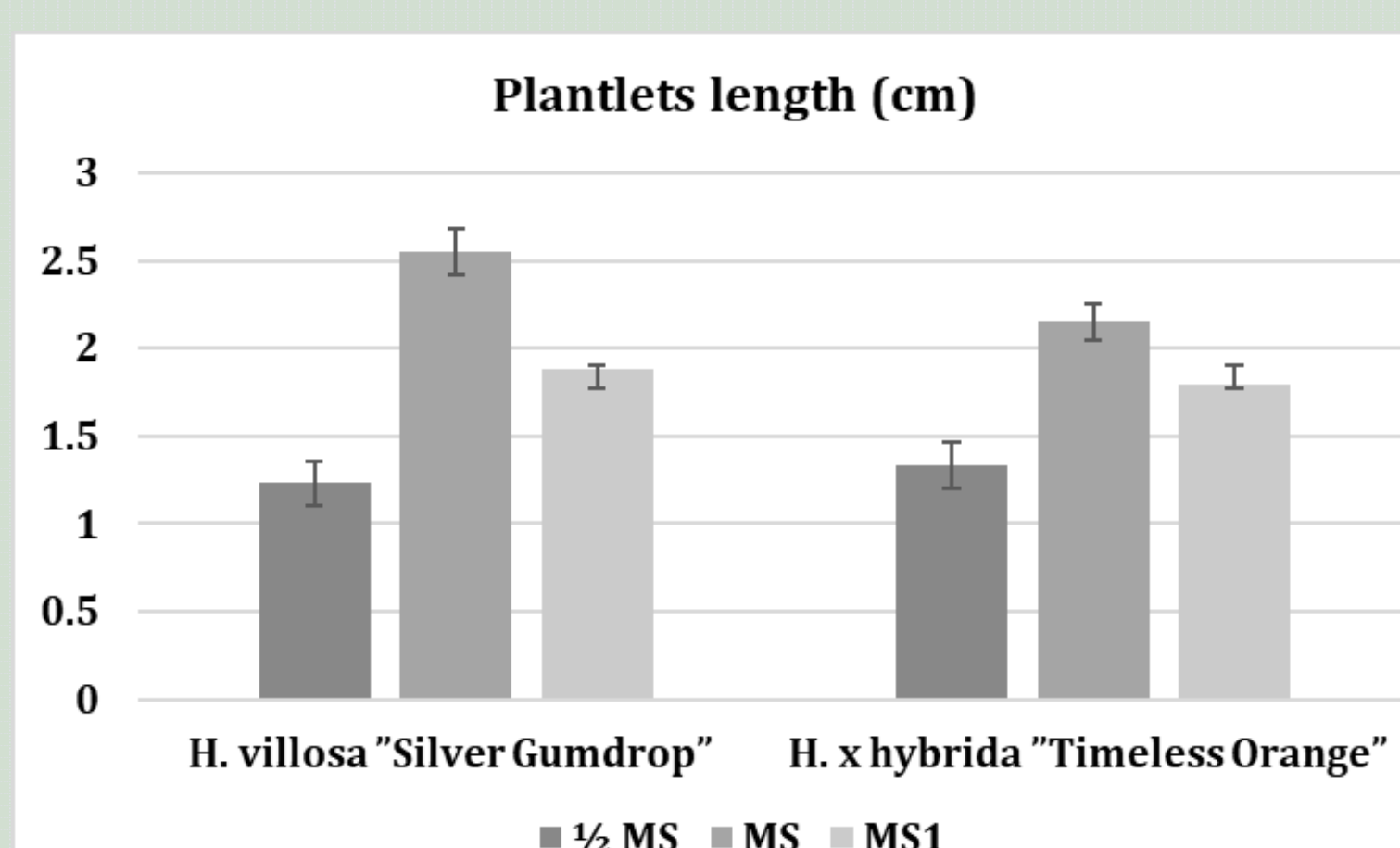


Figure 3. *Heuchera* plantlets after 60 days of culture

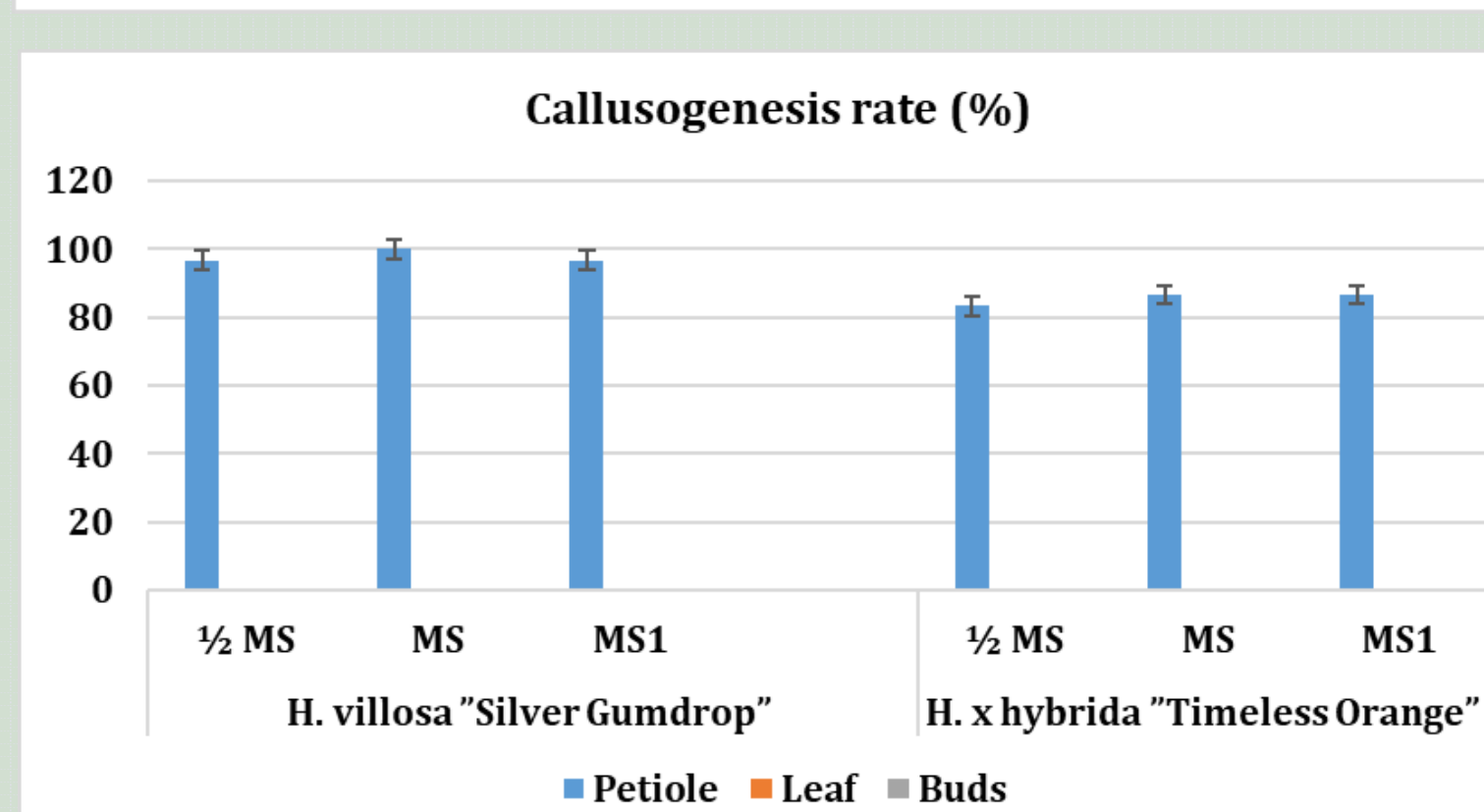


Figure 4. The interaction of genotype, and culture media on plantlets length (cm).

Figure 5. The interaction of genotype, explant type and culture media on callusogenesis rate (%).