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In vitro cultive of cedar with supplementation of the PDA culture media.

G. Marca & T. Gerber

Faculdade Concórdia

Author for correspondence: gerberthaise@gmail.com

Abstract. The presente study had as finality to evaluate the germination of cedar seeds *in vitro* in PDA culture media. The experiment was realized on the Vegetable Biotechnology laboratory of FACC – Faculdade de Concórdia/SC. For the procedure, 200g of english potato were used, put on boiling process for 20 minutes in 1L of distilled water, next, the broth of this solution was used. 20g of saccharose ans 12g of agar were added on it, pH adjusted in 5,8, and 8mL distributed on test tubes, totalizing 40 tubes taken to autoclaving process along with tweezers, distilled water and others, immersed for 75 minutes in sodium hypochlorite (2,5% v/v), and after that the inoculation process on the culture media was realized. The feature evaluated on this experimente was the possible germination of cedar seeds in PDA culture media. The cultures were mainteined on growing room with controled temperature of 25 ± 2ºC, under photoperiod of 16 hours, provided by fluorescente lamps Philips TDL (22.3 µmol.m^{-2}.s^{-1}) and relative humidity of 70%. The results demonstrated 80% of seeds germination. Yet, it's interesting to work with PDA culture media for cedar seeds germination.
Keywords: potato, saccharose, Cedar.

Introduction

The Cedar is a native species from the Atlantic forest which has a big commercial interest, due to itswood used by woodworking, civil construction and aeronauthics. Component of the Meliaceae Family, *Cedrela fissilis is* a Melica specie that can be used in projects of urban afforestation, specially recommended on the recuperation of degraded areas and non-floodable riparian forests (Lorenzi, 2000; Durigan et al., 2002; Xavier et al., 2003). Besides, it presents fast growing (Santos et al., 2001).

The flowering happens from September to December and the fruits mature after the leaves fall, between July and August (Reitz et al., 1983; Carvalho, 1994). An isolated tree can produce more than 1.500 fruits, with more than 60.000 fertile seeds (Rizzini, 1981).

In the production of seedlings by seeds, the fruits should be collected mature directly from the tree, but still closed to prevent the lost of seeds, and after the harvest, taken to complete the dehiscence on a dry and ventilated ambient (LORENZI, 1992). The presence of pathogens after the point of phisiologic mature or seeds storage is a serious threat to quality, due to the fact that high percentages of infected seeds are associated (Yorinori, 1982).

This way, due to the difficulty of cedar seedlings production without the presence of pathogens, this work has as objective to evaluate the germination of cedar *in vitro,* in PDA culture media.

Methods

The experiment was realized at the Vegetal Biotechnology laboratory of FACC – Faculdade de Concórdia/SC. Seeds and Axenic seedlings of *Cedrela fissilis* were used as font of explants for realizing the experiments.

The seeds were previously gotten at Instituto Florestal de São Paulo and collected at Estação Experimental de Araraquara, São Paulo. About 30 seeds were chosen to realize the experiment. After that, the PDA culture media was prepared (potato – dextrose – agar), where 200g of english potato were used, cut in slices of 2cm each, and taken in boiling for 20 minutes in 1L of distilled water. Next it was put 20g of saccharose and 12g of agar, pH adjusted in 5,8, distributing 8 mL on each test tube, totalizing 40 test tubes being taken to autoclaving process, along with tweezers, distilled water and others, in a temperature of 121ºC for 30 minutes.

For the seeds assepsie, they were immersed for 75 minutes in sodium hypochlorite (2,5% v/v). After that, the inoculation process of the seeds was realized in the culture media. The cultures weke kept in the growing room with controled temperature of 25 ± 2ºC, under photoperiod of 16 hours, provided by fluorescent lamps Philips TDL (22.3 µmol.m^{-2}.s^{-1}) and relative humidity of 70%.

Next, the process of disinfection of the seeds happened, where they were washed with distilled water and neutral detergent, rinsed four times and, in laminar flow, they were immersed for 75 minutes in commercial solution of sodium hypochlorite (Q-boa) with 2.5% (v/v) of active chlorine, increased by some drops of detergente. Then, they were rinsed four times with sterelized distilled water, for removing residues of hypochlorite, detergent and inoculated in the culture media (Picture 06) (Nunes *et al.*, 2007).

Next, after the disinfection process, the seeds were inoculated in PDA culture media (Figure 1). The test tubes were covered with polypropylene film (76 mm x 76 mm), attached with rubber bands. The cultures were kept in the growing room with a controlled temperature of 25± 2ºC, under photoperiod of 16 hours, provided by fluorescente lamps Philips TDL (22.3 µmol.m^{-2}.s^{-1}) and humidity of 70% (Nunes *et al.*, 2007), This conditions were used in all of the experiment, except when specified.

The evaluated caracteristic of this experiment was the possible germination of cedar seeds on the PDA culture media.

Figure 1: Inoculated seeds on culture media.

Results and discussion

The results demonstrated that the PDA culture media for germination of cedar seeds *in vitro* was effective. The results showed 80% of seeds germination, as figure 2 presents, showing that in the first days there were no germinative percentage and from the fifth day, the seeds started to uniform the germination process.

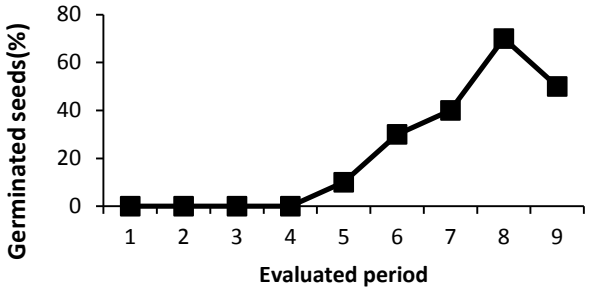

Figure 2: Evaluation of cedar seeds germination at 9 days of growth *in vitro.*

Figure 3 presents the germination process of cedar seeds, forming the seedlings. According to Ferreira (2004) the germination can be defined as the left of the rest condition of the embryo and return of the metabolic activity, also the development of the embryo and emergence of the seedling till it becomes independente from de seeds reserve (Ferreira; Borghetti, 2004; Carvalho; Nakagawa, 2012). Externally it's marked by the forehead's disruption and extrusion of the seedling or primary root (Fenner; Thompson, 2005).

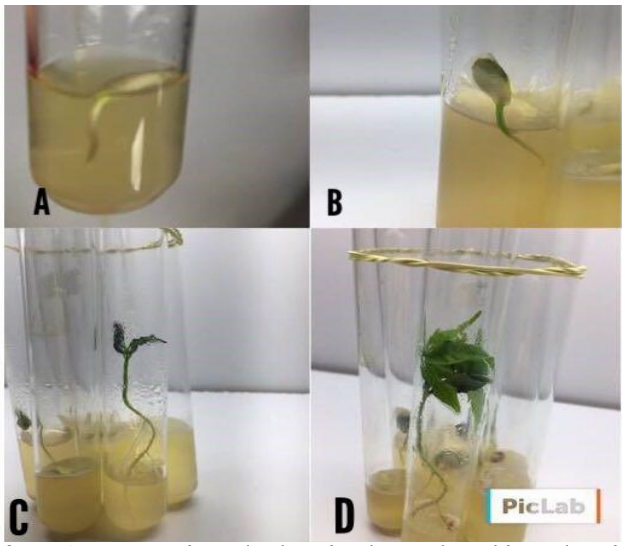

Figure 3: Process of germination of cedar seeds and formation of seedlings. Source: the authors.

There are many factors that influence on the germination process, and they can be classified as intern (instrinsic, as longevity and viability) and extern, associated to climate conditions, as humidity, temperature, light and oxygen. The temperature is one of the main factores that influence the seed, affecting the total germination and the germination speed, because it tends to influence the water absorption speed and the determinant biochemical reactions on the germinative process (Ferreira; Borghetti, 2004; Carvalho; Nakagawa, 2012).

Each specie presents a minimum, maximum and great temperature for germination. The

temperature is called great when it occurs the maximum germination in the minimum time. Above and under the maximum and minimum limits, respectively, the embryons can die. The zone between 20 to 30º is proper for germination of a huge number of subtropical and tropical species (Larcher, 2003; Ferreira; Borghetti, 2004; Brancalion et al., 2010).

Conclusion

As exposed, it can be affirmed that the results gotten with the experiment were positive, and the germination of *in vitro* cedar seeds cultivated in PDA culture media were good. The PDA culture media is an easy and economic way that can produce good seedlings in a bigger scale.

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