CALLUS INDUCTION OF COMMON BEAN (*Phaseolus vulgaris* L.) cv. AZUFRADO HIGUERA

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Introduction. Common bean (*Phaseolus vulgaris* L.) takes an important part of the diet of mexican population. It is a legume widely distributed and available. It has been cultivated from about eight thousand years ago and during that time a great diversity of types and qualities of bean was developed. México has the higher number of species of genus Phaseolus and of wild and creole forms of *Phaseolus vulgaris.* In order to improve its alimentary and agronomic characteristics there are several biotechnological tools like tissue culture.
The aim of this study was to select the best explant and plant growth regulators to establish *in vitro* callus induction of *Phaseolus vulgaris* L. cv. Azufrado Higuera.

Methods. *In vitro* plantlets were obtained from bean seeds. The sterilization protocol was according to Vanegas- Espinoza (1) with some modifications. Percentage of contamination and germination was evaluated. Stem and leaf explants were tested for callus induction on Murashige and Skoog (2) medium added with 6- benzylaminopurine (BAP) 1.5 mg/L and indoleacetic acid (IAA) 0.5 mg/L as reported by Mahamune *et al*.(3). Auxin exchange for 2,4- dichloro phenoxyacetic acid (2,4-D) was evaluated.

Results. Seed germination was 100%, without contamination (Fig. 1A, 1B)

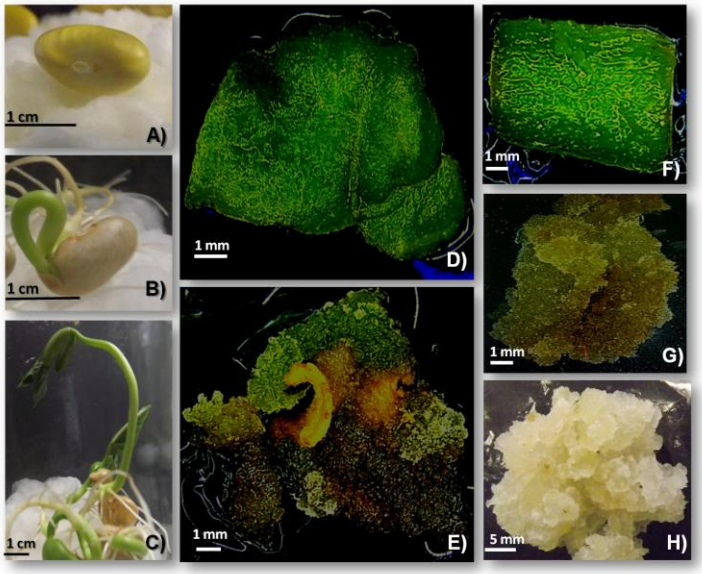

Fig. 1. *Phaseolus vulgaris* L. cv. Azufrado Higuera: A) seed at day 0 B) Seed at day 5 C) Plantlet at day 11 D) Leaf explant with 2,4- D 0.5 mg/L and BAP 1.5 mg/L at day 0 E) Leaf explant with 2,4- D 0.5 mg/L and BAP 1.5 mg/L. at day 16. F) Stem explant 2,4- D 0.5 mg/L and BAP 1.5 mg/L at day 16 G) Callus from stem with 2,4- D 0.5 mg/L and BAP 1.5 mg/L at day 16 H)Callus from stem with 1 mg/L and BAP 0.5 mg/L.

Showing that protocol was appropriate for aseptic *in vitro* culture establishment (Fig 1C). BAP/IAA combination was not good for dedifferentiation induction on both leaf and stem explants of *Phaseolus vulgaris* L. cv. Azufrado Higuera. They turned dark brown and no callus induction was observed. 2, 4- D was evaluated instead of IAA observing that explants (Fig. 1c) started dedifferentiation at day 9. Leaf explants did not dedifferentiate completely and turned brown (Fig. 1D, 1E) and the stem conserved a light green color (Fig. 1F, 1G). Stem explant response to different combinations of BAP and 2, 4-D was evaluated and the best callus was obtained at combination with 1 mg/L 2,4-D and 0.5 mg/L BAP (1H).

Conclusions. The plant growth regulators for callus induction on *Phaseolus vulgaris* L. cv Azufrado Higuera were BAP 0.5 mg/L and 2,4-D 1 mg/L. The best explant for the same aim was the stem. Thereby we have the first step for the establishment of a genetic transformation system of *Phaseolus vulgaris* L. cv. Azufrado Higuera using biolistic.

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