

Characterization of *Phaseolus vulgaris* cell suspension culture

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Stability, as well as intracellular targeting, of normal or modified proteins is being studied by expressing them in transformed plants. This approach can also be used for assessing effectiveness of toxic constituents on crop protection.

For many important crop plants, regeneration protocols from tissues or single cells are not yet routinely available. In this case, the fate of the foreign protein can be followed by transient expression of the gene in a cell system derived from the plant tissue where the gene will be expressed.

To study the expression of modified bean seed proteins we have developed a homologous system easily transformable, based on protoplasts derived from cotyledonary cell suspension cultures (BIC, this issue).

To determine whether these cultures were still expressing major proteins proper of the starting tissue (the developing cotyledon) analysis by Western blot and haemagglutination assay were performed (Bollini et al., *Physiol. Plant.* 65, 15-22, 1985).

Crude extracts of the cultured cotyledons, analysed with immune sera raised against phaseolin and phytohemagglutinin, indicated that during callus induction both proteins gradually disappeared from the tissue.

On the contrary to data obtained from calli derived from maize endosperm (Giovinazzo et al., *Plant Mol. Biol.* 19, 257-263, 1992), the proliferating bean callus did not contain detectable amounts, on Western blots, of both proteins investigated. Absence of phytohemagglutinin was confirmed when haemagglutination tests were carried out directly on crude extracts of both callus and suspension cultures.

These cell cultures therefore represent an appropriate system to study intracellular transport of modified bean proteins for, in addition of being an homologous system, it lacks protein background that could interfere with detection of proteins newly synthesized after transformation.

We are now optimizing culture conditions for PEG-transformed bean protoplasts to obtain stable transgenic calli. This should be a suitable system to investigate toxicity of heterologous proteins against insect larvae.