Abstract of Presentation

Note: This paper should be typed in "Times New Roman" of 12pt.

Name (Underline the family name)
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Presentation Title(Should be no more than 20 words):

Veterinary vaccines produced in plants: First report of a experimental vaccine against BVDV produced in transgenic plants that induce protection in cattle's

Abstract :

Bovine viral diarrhea virus (BVDV) is a pestivirus belonging the *Flaviviridae* family, it is an important cause of mortality, morbility and economic losses of cattle with a worldwide distribution. Subunit vaccines provide the opportunity of developing safe vaccines. However, the challenge is to generate a protective immune response to a cost affordable for veterinary applications.

Transgenic plants for expression of viral and bacterial antigens have been increasingly tested as an alternative methodology for the production of experimental vaccines, based on their scaling-up simplicity, low risk of pathogen contamination and possibility of oral administration. However, a major drawback in most cases is the low expression level of the recombinant antigens in plant tissues, which reduces practical applications. One strategy to solve this problem is the use of strong promoters for conducting transgene expression. Previous results obtained by our group employing transient expression assays in alfalfa (*Medicago sativa*), showed that Cassava Vein Mosaic Virus (CsVMV) promoter presented higher transcriptional activity than the 35S promoter.

Another alternative to overcome the low expression level is to increase the vaccine immunogenicity. A central event in the development of an adaptive immune response is the presentation of the antigens to $CD4^+$ T cells from antigen presenting cells (APCs). In consequence, a way to obtain a more intense specific immune response is to increase the number of MHC-peptide complexes on the surface of APCs. This would be possible by fusing antigens to specific antibodies against APCs´ surface markers.

We have previously reported the production of transgenic alfalfa plants which express a truncated version of BVDV glycoprotein E2, without the transmembran domain (tE2) and fusion protein between tE2 and a single chain antibody against MHCII (ScFv-tE2). Expression of recombinant proteins in these plants was evaluated by Western blot and ELISA. Antigen production was estimated to be approximately 1.5, and 1.7 ugr/gr of leaf for tE2 and ScFv-tE2, respectively. Moreover, binding of ScFV-tE2 to the surface of

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peripheral blood mononuclear cells (PBMCs) from different species was confirmed by flow cytometry.

In this work we optimized the production of recombinant antigen from alfalfa tissues and evaluated immunogenic properties of vaccines formulated with these antigens.

Two methodologies were tested in order to increase recombinant protein production: ultrafiltration by Centrifugal ultrafiltration devices and **ATPS** (aqueous two-phase partitioning system):