

**SUSTAINABLE AND INTEGRATED  
USE OF AGAVE**

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**USO INTEGRAL Y  
SUSTENTABLE DEL AGAVE**



International  
Symposium on Agave

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## Introduction

Studies involving the formation of gametes, double fertilization, and embryo and endosperm development allow the understanding of factors that control the processes of embryo and seed development. In this context, the female gametophyte plays a critical role in every stage of the reproductive process including the transport of sperm nuclei inside the embryo sac (ES) in the process of double fertilization. There is a considerable amount of evidence on the involvement of actin and other motor proteins in the process of elongation of the pollen tube and the movement of sperm nuclei along the pollen tube [1; 2]. There have been some reports of the formation of actin coronas within the ES at the time of double fertilization in plants such as Maize and *Torenia* [3], however, there is no such information in Asparagaceae.

The main purpose of this study was to identify how the actin cytoskeleton is arranged in the mature ES in species of the Asparagaceae family.

## Methods

To The plant material consisted of mature flowers of *Agave tequilana*, *A. salmiana*, *Polianthes tuberosa* and *Manfreda* sp. Ovules of these flowers were dissected and treated either by Feulgen staining [4] which stains DNA and slightly cytoplasm or by staining with rhodamine-phalloidin for detecting actin following [3] with some modifications. The analysis of samples was performed on a Leica TCS SPE confocal microscope. Images were handled with the LAS X software (Leica Microsystems).

## Results and discussion

Feulgen-stained ovules and confocal microscopy revealed an unknown arrangement of strands (TVS) that traversed the central cell vacuole. These strands formed a tunnel-like structure going from the egg apparatus to the central cell nucleus (Fig. 1a). The structural integrity of TVS depends on actin filaments that run inside these cytoplasmic tunnels. Current progress in this research project indicates the presence of putative actin long filaments that traverse the ES (Fig. 1b). It is possible that these actin strands are equivalent to the actin coronas observed in other plant species such as Maize and *Torenia* [3]. The putative actin strands observed in this work might facilitate the movement of sperms inside the ES.

## Conclusions

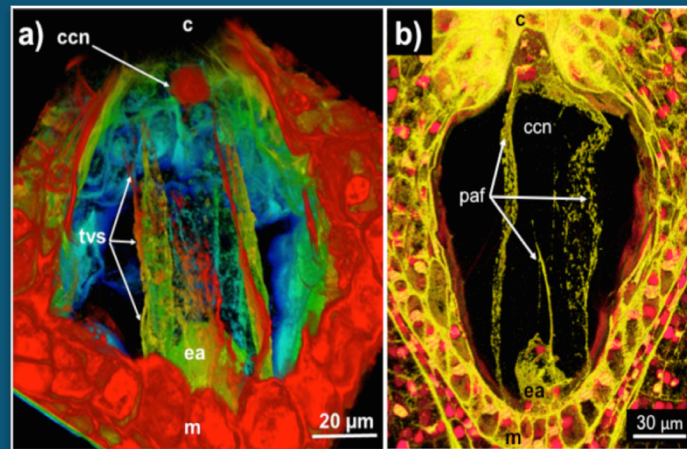
The putative actin strands observed in this work might facilitate the movement of sperms inside the embryo sac.

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**Fig. 1.** a) Mature embryo sac of *P. tuberosa* treated with the Feulgen technique shows strands that traverse the central cell vacuole. b) *A. salmiana* embryo sac stained with rhodamine-phalloidin, showing putative actin filaments aligned from the ccn to the ea. c=chalaza, ccn=central cell nucleus, m=micropyle, tvs=traverse strands, ea=egg apparatus, paf=putative actin filaments.