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Name of the project:
ROLE OF POLYAMINES IN CELL PROLIFERATION, DIFFERENTIATION AND PROGRAMMED CELL DEATH DURING SEED DEVELOPMENT OF SCOTS PINE

The polyamines (PAs) spermidine, spermine and putrescine are wide-spread in higher plants and implicated in various physiological and developmental processes. In plants, PAs are connected to various growth, developmental and stress related processes as well as cell death. It has also been proposed that cell death and cell proliferation are linked and polyamines may provide one possible connection between these contrary phenomenons. In plant cells the most common PAs are putrescine, spermidine, spermine and in some cases cadaverine, which are found as free amines or as amide conjugates.

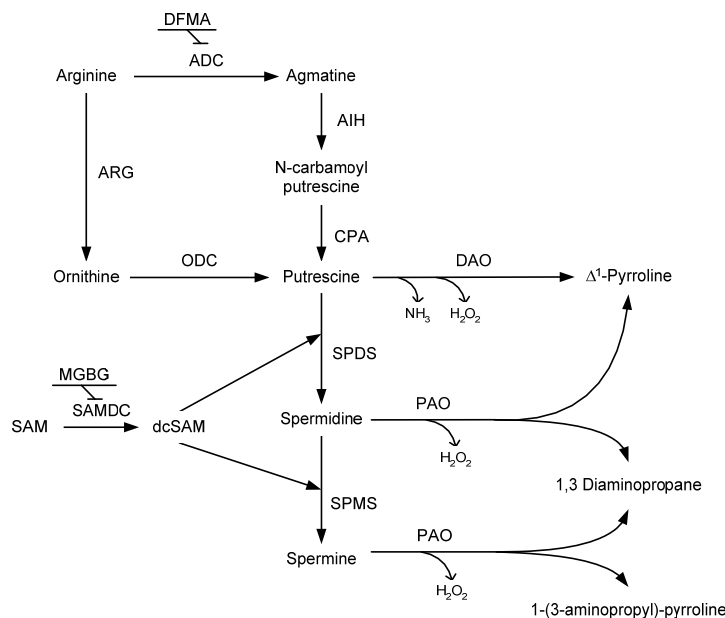


Fig. 1. Pathways of biosynthesis and degradation of the major plant polyamines (putrescine, spermidine and spermine). Enzymes in the pathways: arginase (ARG), arginine decarboxylase (ADC), agmatine imidohydrolase (AIH), N-carbamoylputrescine amidohydrolase (CPA), ornithine decarboxylase (ODC), spermidine synthase (SPDS), spermine synthase (SPMS), diamine oxidase (DAO), polyamine oxidase (PAO) and SAM decarboxylase (SAMDC). The inhibitors of ADC and SAMDC enzymes are α -difluoromethylarginine (DFMA) and methylglyoxyl-bis(guanylylhydrazone) (MGBG), respectively.

Zygotic embryo development and programmed cell death in *Pinus sylvestris*

In gymnosperms, the overall embryo development pathway can be divided in three distinct phases, called proembryogeny, early embryogeny and late embryogeny which culminates with the maturation of the embryo. The pine seed has been proposed to be a model system for studying the eukaryotic PCD machinery but plant seed may also function as a model system when considering the role of a variety of endogenous DNA damaging agents and environmental stresses on genome integrity. This is because from the early stages of development within the mother plant the seed experiences developmentally programmed as well as environmental stresses.

In our recent studies on zygotic embryogenesis of Scots pine (*Pinus sylvestris* L.) we found that both ADC mRNA and ADC protein localized in dividing cells of Scots pine embryo meristems suggesting the essential role of ADC and ADC route in the mitotic cells (Vuosku et al., 2006). We also found that catalase gene expression increased drastically at the late embryogeny stage indicating exposure to oxidative stress. At the same developmental stage, DNA strand breaks accumulated into nuclei in the megagametophyte tissue, but large scale PCD could not be found (Vuosku et al., 2009). We hypothesize that PAs may have important role during embryogenesis in developmentally programmed stress. During late embryogeny H_2O_2 derived from PA oxidation may result in DNA damage and cell cycle arrest. On the other hand PAs may also protect against oxidative stress and prevent large scale PCD. Our ongoing research activities are focused to test the possible role of PAs as bivalent regulators during developmentally programmed stress.

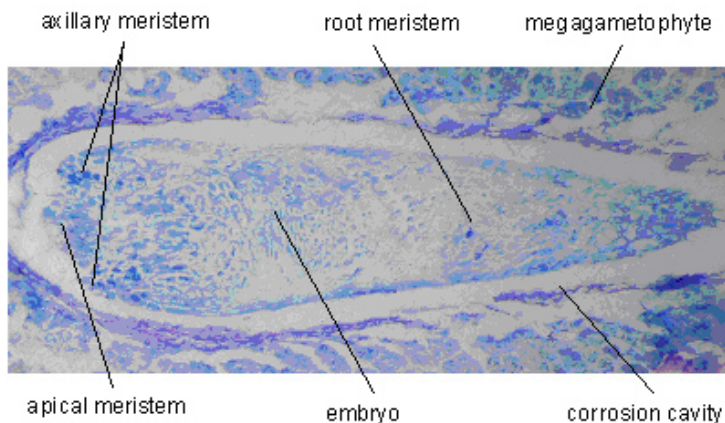


Fig. 2. Scots pine embryo at late embryogeny stage.

Polyamine metabolism and environmental stress

Polyamines play also a key role in stress tolerance. It is known that frequency of diverging temperature and drought stresses in boreal forests has been proposed to increase in future due to climate change. Spd acts indirectly during stress by maintaining high Spm levels while Spm is the direct effector of stress tolerance. Amine oxidases catalyze PA oxidation reaction where hydrogen peroxide, H_2O_2 , is produced. H_2O_2 is involved in the expression of defence and detoxification genes, cell death and developmental programmed cell death.

Our aim is also to study the metabolite and transcriptome level changes in PA-metabolism in Scots pine seedlings prone to especially drought stress.