

The Council for Tobacco Research-U.S.A., Inc.
Review of Grant Application

Grant Application #: 3869

Principal Investigator: Heidi Stuhlmann, Ph.D.

Grant Title: Functional Analysis of Oct-3 During Mammalian
Development

Reviewer: Peter K. Vogt, Ph.D.

Description:

The transcription factor, *Oct-3*, belongs to the POU family of transcriptional regulators. *Oct-3* is expressed in undifferentiated embryonic stem cells, but is invariably down-regulated when these cells are induced to differentiate. It appears that *oct-3* may be necessary to establish and maintain a highly pluripotent phenotype including germ cells, and its down-regulation may be necessary for normal differentiation during embryogenesis. The aims of this proposal are to study the functions of *oct-3*. This will be done by generating retroviral vectors that express *oct-3* cDNA. These vectors will be either defective and, therefore, will not produce infectious viral progeny, or they will be replication competent and synthesize infectious progeny. The function of *oct-3* will be explored in the following ways: (1) By infecting ES cells and then studying their abilities to differentiate *in vivo* as part of a mosaic embryo or *in vitro* in the form of embryoid bodies. (2) The function of *oct-3* will also be examined by infecting post-implantation mouse embryos with the *oct-3* expressing vectors. In preliminary studies, vectors have been constructed both of the replication-defective and replication-competent type. They produce *oct-3* in its native form and in a variant form that is tagged with an 18-amino acid epitope from the influenza hemagglutinin. The vectors appear to be reasonably stable. They are produced in comparatively low titers, but titers that are sufficient to achieve the infection of ES cells and the infection of post-implantation embryos. So the tools are at hand for an ectopic expression of *oct-3* and for an analysis of the phenotypes that such an expression produces.

Critique:

Transcription factors which control early development and which play a role in maintaining a pluripotent phenotype are important. *Oct-3* may belong to this category as judged from its preferential expression in undifferentiated ES cells and in primordial germ cells. This proposal is well focused. It is based on a considerable amount of careful and successful preliminary work and it takes advantage of the skills in mammalian embryogenesis that the applicant has developed during her postdoctoral studies.

Investigator:

Heidi Stuhlmann has a Ph.D. from the University of Hamburg. She worked at the Whitehead Institute with Richard Mulligan and Rudi Jaenisch, and at Stanford with Paul Berg. She is clearly both a seasoned embryologist and virologist at the same time with an outstanding record of publications.

Budget:

The budget is remarkably modest and she does not have a great deal of other support.

Recommendation:

Recommendation is for approval with high priority (1.0 to 1.2).