ABSTRACT
DEPLETION OF THE RNA HELICASE SKIV2L2
IMPEDES MITOTIC PROGRESSION AND
HISTONE mRNA TURNOVER IN
MURINE CELL LINES

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RNA surveillance via the nuclear exosome requires cofactors such as the helicase
SKIV2L2 to process and degrade certain noncoding RNAs. This dissertation characterizes the
phenotype associated with RNAi knockdown of SKIV2L2 in two murine cancer cell lines:
Neuro2A and P19. Skiv2l2 knockdown in Neuro2A and P19 cells induced changes in gene
expression indicative of cell differentiation and reduced cellular proliferation. Analysis of the
cell cycle revealed defective progression through mitosis following SKIV2L2 depletion. These
results suggest that SKIV2L2 enhances mitotic progression, thereby maintaining cancer cell
proliferation and preventing differentiation. Indeed, SKIV2L2 levels were found to be
downregulated during chemically induced differentiation, further implicating SKIV2L2 in
maintaining proliferation and multipotency in cancer cell lines.

Since SKIV2L2 targets RNAs for processing and degradation via the nuclear exosome, it
was hypothesized that with SKIV2L2 depletion, the accumulation of some RNA target triggers
mitotic arrest. In search of such a target, RNA-seq was utilized to identify RNAs that were
elevated in Skiv2l2 knockdown cells. SKIV2L2 depletion resulted in the accumulation of non-
coding RNAs, intergenic RNAs, ribosomal protein mRNAs, and replication-dependent histone
mRNAs. Since the regulation of histone mRNAs is tightly linked to the cell cycle, further
experiments were conducted to confirm SKIV2L2 targets histone mRNAs for degradation. RNA
immunoprecipitation demonstrated direct binding between SKIV2L2 and histone mRNAs, and
RNA degradation assays showed that the half-life of histone mRNAs doubles with SKIV2L2
depletion. The resulting histone imbalance following loss of SKIV2L2-directed RNA
surveillance could impede mitotic progression, resulting in mitotic defects and indirectly
triggering differentiation.