Summary 96

## 7 SUMMARY

RNA editing in mitochondrial and plastidic transcripts of plants results in the modification of cytidines to uridines (C to U). In vivo, in organello and in vitro systems have been already established to better understand this phenomenon and to identify factors involved in this modification reaction. Nevertheless, they are either time consuming or show only low editing activity so that it is very important to have a very sensitive detection system for investigating RNA editing in vitro. The here developed assay, the primer ligation, can detect less than 1 % C to U editing. It is based on the annealing of two oligonucleotides to the editing substrate and a following ligation if perfectly matched at the editing site. Via this method reliable mitochondrial and plastidic in vitro RNA editing systems could be established with editing activities of up to 5%. The mitochondrial pea system showed heterologous RNA editing at the conserved sites of the atp9 RNA, as did the mitochondria from potato in an additionally established in organello system. Furthermore, for this mitochondrial RNA organellheterologous editing also could be shown in a pea plastidic in vitro system suggesting that not only mitochondria but also chloroplasts contain all essential factors for the atp9 RNA editing.

To exactly determine the sequence requirements in the editing substrate a competition analysis was developed which uses the hybridization of deoxynucleotide competitors to the RNA to inhibit the interaction with possible editing factors. With the help of this method an essential *cis*-element for *atp9* RNA editing was identified for the first time in the upstream region of the editing site 1. Via linker scanning mutagenesis the position of this 10 nucleotide spanning element could be confirmed. Furthermore, the analysis of the insertion mutants showed in *in vitro* as well as in *in organello* systems that the distance (spacing) of the element to the editing site 1 is essential whereas the C to U conversion at the editing site 2 is performed independently.

In this work the foundation has been laid for elucidating the regulation of RNA editing by identifying *cis*-elements and possible *trans*-factors and to better understand the origin of this still enigmatic process.