SYNOPSIS

The life cycle in flowering plants can be divided into two phases: the vegetative and the reproductive phase. The Shoot Apical Meristem (SAM) is a small group of undifferentiated and actively dividing cells at the shoot apex that gives rise to the aerial part of the plant. During the vegetative phase of plant development, the SAM produces leaves in a spiral fashion (in Arabidopsis). When the plant transits into the reproductive phase, the developmental fate of the SAM changes and it produces flowers in place of leaves. The timing of this transition is crucial for maximum reproductive fitness. In higher plants, multiple factors regulate this switch from the vegetative to the reproductive phase, which ultimately leads to the formation of haploid gametes and fusion of the two opposite gametes to form a new organism.

This study is an attempt to understand the role of the Arabidopsis Mei2 Like (AML) genes during plant development, especially their function during reproductive development. These genes code for RNA binding proteins, which show homology to the Mei2 gene from S. pombe. Mei2p is a key regulator of the initiation of meiosis. It is involved in the regulation of pre-meiotic DNA synthesis and entry of the cell into meiosis I. AML like genes are also present in other plant species and show high conservation in the RNA recognition motifs (RRM). In the light of these facts, the AML genes are potential candidates for having a role in plant development, especially reproduction. The work presented in this thesis explores the function of the AML genes and the issue of whether sequence similarity with yeast Mei2 extends to conservation of the biological function.

Chapter 1 is a brief review of the factors regulating reproductive development in plants. First I have summarised various factors that affect the vegetative to reproductive phase transition in lower plants, algae, and ferns. This is followed by an overview of the genetic and molecular mechanisms regulating the floral transition in higher plants. In this section, I have discussed the multifactorial nature of regulation of the floral transition, elaborating on the various pathways repressing or promoting this transition. Meiosis and syngamy form a crucial part of
the reproductive cycle. Towards the end of the introduction I discuss the mechanisms underlying meiosis in plants.

In Chapter 2, the first chapter of the results I present evidence that there is a tight correlation between the induction of the AML genes and the entry of plant into the reproductive phase. Accelerating or delaying the floral transition using different photoperiods, treatment with flowering promoting phytohormone (GA), and flowering time mutants, alters the timing of induction of the AML genes. Under all the different conditions tested the time of increase in the AML transcripts corresponded to the time of flowering. These observations provide strong evidence to support the hypothesis that the AML genes are targets of the flowering time programme. I further show that the induction of these genes in the leaves is independent of signalling from the shoot apical meristem.

Towards the aim of understanding the role of AML genes I have characterized the spatial and temporal expression of these genes using in situ hybridizations and AML3 promoter GUS fusions. Chapter 3 discusses the expression pattern observed for two representative members of the family (AML2 and AML3). The results presented in this chapter point to an association of AML2 and AML3 expression with the actively dividing cells in the vegetative as well as the reproductive phase. These genes are abundantly expressed in the actively proliferating meristematic regions: shoot apical meristem, root apical meristem, and primary and secondary inflorescence meristem. Strong expression of these genes was also detected during male and female gametophyte development (sporogenesis and gametogenesis). Taken together these results show a correlation between cell proliferation and AML expression.

In Chapter 4 I present results from the reverse genetic approaches that I used to understand the function of the AML gene family. I have analyzed mutants carrying T-DNA insertions in individual members of the AML family. The single mutants do not show any defects in vegetative or reproductive development. Of all the double mutants examined the aml1 aml5 combination shows defects in leaf primordia initiation and leaf morphology. These double mutants show altered phyllotaxy. The leaf lamina in these mutants fails to expand and some leaves even
show radialization. These results suggest an involvement of the AML1 and AML5 gene products in determining the position of leaf initiation on the SAM and proper development of the leaf. In addition the results from the anti-sense and the RNAi approaches used to potentially target and down regulate the expression of all the members of the AML family, suggest a role of these genes in male and female gametophyte development. These functions are consistent with the expression pattern observed during expression analysis.

In Chapter 5, I summarize all the results from this work. There is a growing body of evidence that support the crucial role of the RNA binding proteins during various processes of plant development. In this chapter I speculate on the potential mode of action of the AML1 and AML5 genes in regulating leaf initiation and development.

At the end of the report Chapter 6 describes the techniques used during the course of this study.

Taken together the results from this study suggest a broad role of the AML genes in plant growth. During vegetative growth of the plant AML1 and AML5 are involved in maintaining the architecture of the plant. In the reproductive phase these genes appear to be autonomous targets of the flowering time programme in the leaves. In addition, these genes are involved in male and female gametophyte development.