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# Unravelling Epigenetic Dynamics in *Spirodela polyrhiza*: Insights into Transposable Element Regulation

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## Abstract

In flowering plants, the regulation of transposable elements (TEs) involves various molecular processes that lead to the formation of heterochromatin. This state is characterized by heightened DNA methylation (mDNA) and specific histone modifications (e.g. H3K9me2). To ensure TE specificity, 24nt small interfering RNAs (siRNAs) guide mDNA deposition through the RNA-directed DNA methylation (RdDM) pathway. Once established, mDNA can persist independently of siRNAs due to a positive feedback loop with H3K9me2. In contrast to the widespread occurrence of RdDM in the vegetative and sexual tissues of flowering plants, *Spirodela polyrhiza*, a member of the Lemnaceae family, exhibits a distinct pattern. This aquatic species displays reduced mDNA, minimal RdDM expression, and a near absence of 24-nt siRNAs during clonal vegetative reproduction. Furthermore, some key components of RdDM, mDNA maintenance and RNA silencing are conspicuously absent from its genome. The characterization of the TE epigenetic landscape in *Spirodela* reveals a unique pattern where the loss of mDNA and H3K9me2 coincides with TE decay in gene-rich regions. However, remnants of TEs persist in a silenced state, marked by H3K9me1. In contrast, the few intact TEs exhibit significant DNA methylation, H3K9me2, and, interestingly, 21- and 22-nt siRNAs, resembling the patterns observed in TEs subjected to RdDM in other angiosperms. Notably, *Spirodela* produces 22-nt siRNAs from transiently expressed double-stranded RNA, even in the absence of DCL2, an siRNA production-associated enzyme. While unlikely involved in RdDM, this suggests an alternative silencing pathway in *Spirodela*, possibly subject to tissue or developmental regulation. These findings underscore the importance of diverse plant models in elucidating silencing pathway complexities, enhancing our understanding of plant epigenetics.

**Keywords:** transposon, RdDM, mDNA, chromatin, *Spirodela*

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