

The spermatogenic potential of the volvocine algal sex determining gene *MID* evolved prior to dimorphic sexes

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Abstract

Volvocine algae comprise a unique comparative model for investigating the evolution of distinct male and female sexes (anisogamy/oogamy) from an isogamous ancestral state with mating types. In *Chlamydomonas reinhardtii* a single *minus* mating (*MT*-) type gene called *CrMID* (minus dominance) determines mating-type. Mid is putative RWP-RK family transcription factor and orthologs of Mid are present throughout the volvocine algal lineage in either the *MT*- or male mating locus of each species. In *Chlamydomonas* ectopic expression of *CrMID* in a *MT*+ strain (*MT*+::*CrMID*-*T*) causes sexual differentiation with a *minus* phenotype, while in the multicellular species *Volvox carteri* ectopic expression of the male *MID* gene (*VcMID*) in females (*Eve*::*VcMID*-*T*) is sufficient to induce spermatogenesis (Geng S. et al 2014); but the *VcMID* and *CrMID* genes could not induce ectopic sexual differentiation when expressed heterologously in *Chlamydomonas* (*MT*+::*VcMID*-*T*) or *Volvox* (*Eve*::*CrMID*-*T*) respectively. Thus, the function of the Mid protein and/or its associated network in sexual differentiation has evolved in the volvocine lineage. We used ectopic cross-species expression experiments with *MID* genes from different colonial volvocine genera to identify when it acquired the ability to induce sperm development. Reciprocally, we also tested whether ectopically expressed *MID* genes from colonial Volvocine species could function in *C. reinhardtii* to control *minus* mating type differentiation. We expressed epitope-tagged *MID* genes from isogamous (*C. reinhardtii*, *G. pectorale*) or oogamous (*P. starrii*, *V. carteri*) volvocine species in *V. carteri* female strain Eve and tested for their ability to induce spermatogenesis. Transgenic female *V. carteri* expressing *PsMID* produced functional sperm packets during sexual development, which is similar to *Eve*::*VcMID*-*T*, but with slightly lower efficiency (95% versus 100% sperm packets). The sperm packets from *Eve*::*PsMID*-*T* strains also had hatching defects that were more severe than in *Eve*::*VcMID*-*T* strains. Transgenic female *V. carteri* expressing *GpMID* had a more complex phenotype, with smaller vegetative spheroids and a disorganized pattern of somatic cells compared with controls. Remarkably, when sexually induced, *Eve*::*GpMID*-*T* strains produced self-fertile hermaphrodites with mixtures of sperm packets and eggs within a single parental spheroid. It is somewhat paradoxical that the spermatogenic potential of Mid evolved in the isogamous species *Gonium pectorale* prior to the evolution of oogamy. This finding suggests that changes in the cis-regulatory networks controlled by Mid proteins rather than changes in Mid sequence were responsible for innovations leading to the emergence of anisogam/oogamy.

Reference

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