

Oogenesis: Making the Mos of Meiosis

Meiosis is an ancient type of cell division whose advent allowed the evolution of sexual reproduction. The evolutionary history of the specialization that allowed gamete production to emerge from a simple reduction division has been unclear. New data now suggest that the molecular mechanisms involved in animal oocyte specialization may have origins that predate the emergence of bilaterian animals.

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There is perhaps no single subject as fascinating to biologists as sex. As with all fundamental biological questions, the evolutionary origins of sex have long been the subject of both theoretical and empirical work [1]. The chief selective advantage of sexual reproduction is thought to be increased allelic variation, a result both of meiotic crossing over and of the fusion of haploid genomes (see, for example, [2]). However, both asexual and parthenogenetic reproduction have clearly evolved independently multiple times in animals, and both constitute successful reproductive modes [3]. Although there is no single current model that entirely accounts for the ancient origins and persistence of sex, one key cellular process distinguishes gametogenic cell divisions from clonal ones: meiosis.

Although not universal among extant eukaryotes, meiosis appeared early in eukaryotic evolution [4]. It evolved from a pre-existing mitotic program, and the core genetic machinery for meiosis was probably present in single-celled eukaryotes [5]. Oocytes of sexually reproducing animals must (1) be haploid; (2) be fertilizable; and (3) not begin the mitotic divisions of embryogenesis before fertilization has taken place. The latter two features are critical components of oocyte maturation and are ancient characteristics common to oocytes from all metazoan branches. Because the timing of oocyte maturation within the meiotic cycle is variable (reviewed in [6]), it has been unclear to what extent the molecular mechanisms underlying this critical aspect of meiosis are evolutionarily conserved. A recent paper in *Current Biology* by Amiel and colleagues [7] has uncovered a conserved role for a germ-cell-specific kinase in cnidarian and vertebrate oocyte maturation. Their results

suggest that, although meiotic maturation can occur at different times during meiosis, some of its important features share common, ancient molecular mechanisms.

All oocytes pass through two divisions, meiosis I and meiosis II (Figure 1). Prophase I of meiosis I is followed by germinal vesicle breakdown (GVBD), and between these two events, many oocytes arrest the progression of meiosis. This primary arrest may last for hours to years and can be released by fertilization or by hormonal triggers, as first shown by Heilbrunn and colleagues in 1939 [8]. In some animals, a second arrest period also occurs between metaphase of meiosis I (MI) and completion of metaphase of meiosis II (MII). This secondary arrest is generally released by fertilization. Landmark experiments published in 1971 identified chemical activities that trigger passage through both of these critical arrest stages [9,10]. Maturation promoting factor (MPF) [9,10] releases primary arrest, while cytostatic factor (CSF) induces secondary arrest and can even block the mitotic division of early cleavage stage embryos [9]. One gene involved in both release of primary

arrest and establishment of secondary arrest is the germ-cell-specific kinase *c-mos*.

Overexpression of *c-mos* in somatic cells results in overproliferation, which led to its initial identification as the first cellular proto-oncogene [11]. The capacity of *c-mos* to release somatic cells from mitotic arrest [12] parallels its activity in oocyte primary arrest release and MPF activity [13]. However, when overexpressed in cleaving blastomeres of early frog embryos, *c-mos* can arrest cell division [14]. This phenotype is reminiscent of its CSF activity, whereby it prevents oocytes from entering premature mitotic division [15]. In vertebrate gametes, *c-mos* homologues participate in both primary and secondary arrest: during MI arrest release, *c-mos* activates a MAPK cascade to effect GVBD, spindle formation, and first polar body formation, and during MII cytostatic arrest, *c-mos* prevents parthenogenetic cleavage divisions in oocytes [12,16].

Studies in mice, frogs, and starfish have shown that *c-mos*' coding sequence, functional roles, and transduction via the MAPK cascade are highly conserved in deuterostomes (Figure 2). However, little is known about its possible roles in protostomes or outside of the Bilateria. In cnidarians, the MAPK cascade is implicated in oocyte maturation, but its upstream activators have remained unknown [17]. Amiel and colleagues [7] searched several metazoan genomes for *c-mos* homologues and found at least one *c-mos* homologue with a conserved

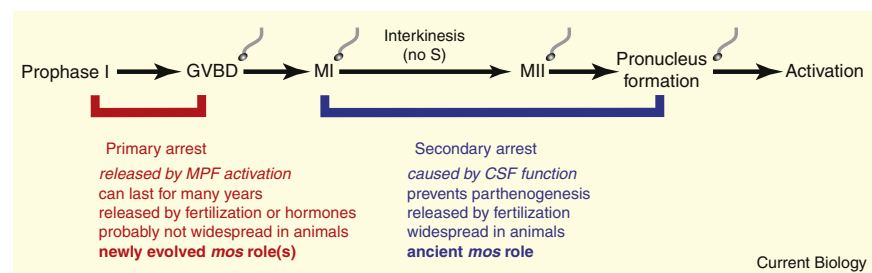


Figure 1. Arrest and maturation points during meiosis.

Progression through meiosis in animals includes up to two arrest periods. Primary arrest (red) occurs after prophase I, but before GVBD, and is released by MPF activity. Secondary arrest (blue) occurs after metaphase I (MI), and before final egg activation, and is induced by CSF activity. Fertilization (grey sperm) can occur at different points of the meiotic cycle in different animals but is always after primary arrest. Given the different activities of *c-mos* homologues across the animal kingdom (Figure 2), the CSF role of *c-mos* may be ancient, while its involvement in primary arrest may be more recently evolved.

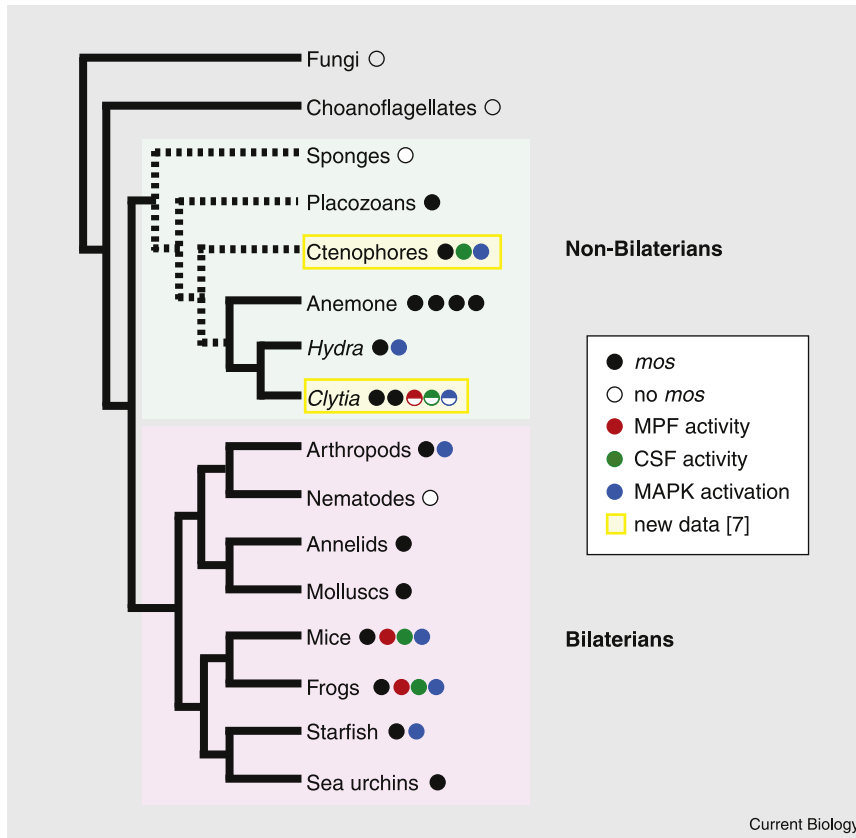


Figure 2. Phylogenetic distribution of *mos* homologues and activities in oocyte maturation. *mos* homologues (black circles) are found in genomes of both bilaterians (pink box) and non-bilaterians (blue box), but not in genomes of choanoflagellates or fungi (white circles). The study by Amiel and colleagues [7] has increased our understanding of the roles of non-bilaterian *mos* genes in oogenesis (yellow boxes). MAPK cascade activity (blue circles) and CSF activity (green circles) are aspects of metazoan meiotic maturation, whereas MPF activity (red circles) may be a more recently evolved role of *mos* that is nonetheless present in some cnidarians. The hydrozoan *Clytia* examined by Amiel and colleagues [7] has two *mos* genes (black circles), one of which has significantly more activity in meiotic maturation than the other (half white circles). The phylogenetic position of some bilaterian outgroups is still controversial (dotted lines) [19,20], which impacts on our interpretation of the evolutionary origins of *mos* genes.

kinase domain in all metazoan genomes except for nematodes. At the base of the animal tree, however, cnidarian, ctenophore and placozoan genomes contain *c-mos* homologues, but the sponge, choanoflagellate and fungal genomes examined do not (Figure 2). Protists do possess homologues of some metazoan ‘meiosis genes’ [18], but the absence of *c-mos* in their genomes may help explain why fungi and choanoflagellates, though capable of reduction divisions, do not generate haploid ‘gametes’ with the carefully controlled cell-cycle progression characteristic of metazoan oocytes. A recent and controversial phylogenetic placement of ctenophores as basal to sponges [19] would support the interpretation that *c-mos* was present

in a metazoan ancestor and subsequently lost in sponges. However, if we interpret Amiel and colleagues’ data [7] using traditionally supported animal phylogenies that place sponges at the base of the metazoan tree [20], *c-mos* was absent from sponge genomes, and first arose in a eumetazoan last common ancestor (Figure 2). Unfortunately, too little is known about sponge oogenesis for us to be able to speculate as to why *c-mos* is missing from, for example, the *Amphimedon* sponge genome.

While a single copy of *c-mos* is present in most animal genomes, the two cnidarian genomes examined have duplicated this locus: the sea anemone *Nematostella vectensis* has

four copies, and the hydrozoan *Clytia hemisphaerica* has two. Both *Clytia* genes (*CheMos1* and *CheMos2*) are similar to their deuterostome orthologues with respect to both gonad-specific expression in both males and females, and kinase activity that results in MAPK cascade activation. The two *Clytia* genes also arrest embryonic cleavages in both frog and *Clytia* embryos, and induce maturation of late-stage (post-MI) frog oocytes. Finally, *c-mos* prevents GVBD and ovulation in early-stage (pre-MI) *Clytia* oocytes. These cnidarian *c-mos* genes are therefore capable of the same biological activities as vertebrate *c-mos*.

Morpholino-mediated knockdown of the two *CheMos* genes demonstrates that, in *Clytia*, the roles of the two orthologues may not be identical. In general, one gene (*CheMos1*) has more detectable kinase activity, and a more severe phenotype than the other (*CheMos2*) with respect to MAPK activation, oocyte maturation induction, embryonic cleavage arrest, and microtubule organization. Oocytes without *CheMos1* activity can progress through GVBD but cannot emit polar bodies, nor do they display cytotstatic arrest. Instead, they undergo disorganized cortical contractions and occasionally even progress to parthenogenetic cleavage. Drug-mediated knockdown of the MAPK pathway phenocopies these *CheMos1* oocyte effects. Both *CheMos* knockdowns and chemical MAPK inhibition result in abnormal microtubule organization, consistent with the premature cortical cytokinetic movements and absence of polar bodies observed in these oocytes.

In summary, while ‘core meiotic genes’ are widespread in metazoans and fungi, *mos* genes are specific to eumetazoans. Both cnidarian and deuterostome *mos* genes have conserved roles in oogenesis and meiosis, and both roles are mediated by MAPK activation. The MAPK-mediated effects of *mos* on oocyte actin and microtubule dynamics are associated with two oocyte-specific events, polar body emission and cytotstatic arrest. The latter may have been of significant adaptive value in the evolution of broadcast spawning, in which sexually mature animals in operatic

habitats release eggs and sperm into the water column, and fertilization takes place between free-floating gametes. This mode of sexual reproduction is exhibited by many extant marine invertebrates and could plausibly have been used by ancestral metazoans. The evolution of cytostatic arrest could have allowed oocytes to be produced and released independently of fertilization, without running the risk of premature parthenogenetic cleavage.

Several outstanding questions remain when considering the evolution of gametogenesis and the roles of *mos* genes in this process: why were multiple copies of *mos* retained in cnidarians, while bilaterians kept only one? Does the absence of *mos* in the sponge genome correspond with a lack of cytostatic arrest in sponge oocytes? Similarly, do eukaryotic protists undergo parasexual cleavages without any arrest phases, in accordance with the absence of *mos* homologues? Are choanoflagellates, lacking *mos*, capable of any kind of reduction division? While answers to these questions will undoubtedly require many more years to achieve, we can now begin to speculate as to the relative evolutionary histories of the many roles of *c-mos*: the meiotic maturation function shared by *Clytia* and deuterostomes suggest that the secondary arrest role of *c-mos* is an ancient one, while its

primary arrest role may be more recently evolved.

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Olfaction: Chemical Signposts along the Silk Road

A recent study on the reception of olfactory cues by silkworm larvae illustrates how the convergence of genomic, physiological and ecological data promises to shed light on the origins and evolution of chemically mediated interactions between plants and insects.

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For insects, olfaction is the primary sensory modality used to acquire information about the world [1]. Information obtained from the detection of odor cues helps insects to negotiate complex environments,

locate valuable resources, and avoid toxic or otherwise life-threatening conditions. Among the most important of these cues are pheromones emitted by conspecific individuals, which convey socially relevant information — for example, by signaling the presence of potential mates. But insects also detect and

respond to other ambient odors that can provide valuable information about local conditions. Foremost in significance among these ‘general odorants’ are volatile compounds emitted by plants. Plants dominate the biomass of terrestrial ecosystems and engage in continuous gas exchange with the surrounding atmosphere, creating a rich olfactory landscape for other organisms. Moreover, the composition of the volatile blend released by plants differs among species and also varies systematically in response to a wide array of environmental conditions, including insect feeding and pathogen infection [2]. As a result, volatile cues can potentially provide foraging insects