

Postovulatory Aging *in vitro* Leads to Maturation Promoting Factor Destabilization and Abortive Spontaneous Egg Activation in Rat

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Abstract—In mammals, freshly ovulated eggs are generally arrested at metaphase-II (M-II) stage of meiotic cell cycle and possess first polar body (PB-I). Culture of these eggs *in vitro* leads to postovulatory aging-induced abortive spontaneous egg activation (SEA). The morphological, biochemical and molecular changes during postovulatory aging-induced abortive SEA remains poorly understood. Hence, the present study was designed to investigate morphological, biochemical and molecular changes during postovulatory egg aging *in vitro*. For this purpose, eggs collected after 14 h of post hCG surge were cultured for 1, 2, 3, 5 and 7 h *in vitro*. The morphological changes, percentage of abortive SEA, phosphorylation status of Cdk1, levels of cyclin B1, Wee-1 kinase, Emi2, APC/C and MAD2 were analyzed. Data of the present study suggest that postovulatory aging triggered initiation of second polar body extrusion, a morphological feature of abortive SEA in a time-dependent manner. Postovulatory egg aging *in vitro* increased Wee 1 kinase as well as Thr-14/Tyr-15 phosphorylated Cdk1 levels, while Thr-161 phosphorylated Cdk1 and cyclin B1 levels were significantly reduced. The Emi2 level was reduced but APC/C and MAD2 levels were first increased significantly and then decreased during *in vitro* egg aging. These results suggest that increased Wee 1 kinase level modulated specific phosphorylation status of Cdk1 that could have resulted in the dissociation of MPF heterodimer. At the same time, decreased Emi2 triggered APC/C activation, which induced cyclin B1 degradation thereby MPF destabilization. The destabilized MPF results postovulatory aging-induced abortive SEA in rat eggs cultured *in vitro*.

Keywords: Postovulatory aging, abortive SEA, Wee 1, Cdk1/ cyclin B1, Emi2/APC/C.