Catechol-O-methyltransferase and methoxyestradiols participate in the intraoviductal nongenomic pathway through which estradiol accelerates egg transport in cycling rats

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Abstract

Estradiol (E2) accelerates oviductal egg transport through intraoviductal nongenomic pathways in cyclic rats and through genomic pathways in pregnant rats. This shift in pathways, which we have provisionally designated as intracellular path shifting (IPS), is caused by mating-associated signals and represents a novel and hitherto unrecognized phenomenon. The mechanism underlying IPS is currently under investigation. Using microarray analysis, we identified several genes the expression levels of which changed in the rat oviduct within 6 hours of mating. Among these genes, the mRNA level for the enzyme catechol-O-methyltransferase (COMT), which produces methoxyestradiols from hydroxyestradiols, decreased 6-fold, as confirmed by real-time PCR. O-methylation of 2-hydroxyestradiol was up to 4-fold higher in oviductal protein extracts from cyclic rats than from pregnant rats and was blocked by OR486, which is a selective inhibitor of COMT. The levels in the rat oviduct of mRNA and protein for cytochrome P450 isoforms 1A1 and 1B1, which form hydroxyestradiols, were detected by RT-PCR and Western blotting. We explored whether methoxyestradiols participate in the pathways involved in E2-accelerated egg transport. Intrabursal application of OR486 prevented E2 from accelerating egg transport in cyclic rats but not in pregnant rats, whereas 2-methoxyestradiol (2ME) and 4-methoxyestradiol mimicked the effect of E2 on egg transport in cyclic rats but not in pregnant rats. The effect of 2ME on egg transport was blocked by intrabursal administration of the protein kinase inhibitor H-89 or the antiestrogen ICI 182780, but not by actinomycin D or OR486. We conclude that in the absence of mating, COMT-mediated formation of methoxyestradiols in the oviduct is essential for the nongenomic pathway through which E2 accelerates egg transport in the rat oviduct. Yet unidentified mating-associated signals, which act directly on oviductal cells, shut down the E2 nongenomic signaling pathway upstream and downstream of methoxyestradiols. These findings highlight a physiological role for methoxyestradiols in the female genital tract, thereby confirming the occurrence of and providing a partial explanation for the mechanism underlying IPS.

Keywords: 17b-estradiol, catechol-O-methyltransferase, estrogen receptor, methoxyestradiols, oocyte transport, oviduct, ovum pick-up/transport