COMMENTARY

Understanding variability in ethanol teratogenicity

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Ethanol is the most widely used psychoactive drug and, when used inappropriately, can have deleterious effects. The developing fetus is particularly sensitive to the deleterious effects of ethanol. Fetal alcohol exposure causes Fetal Alcohol Spectrum Disorders (FASD), which have an estimated prevalence reaching 1% in North America (1). FASD are associated with defects to numerous organs, including the central nervous system (2). Defects to the central nervous system are especially diffuse and can include disruptions to the corpus callosum (3, 4). This variability is problematic for identifying and providing treatments for FASD, making a more complete understanding of FASD variation essential.

Extensive research has gone into understanding the cause of variability in outcomes resulting from embryonic ethanol exposure. The dosage and timing of ethanol exposure are important variables that correlate with phenotypic outcome (5). In addition, genetic influences alter the outcome of embryonic ethanol exposure. In human twin studies, 100% of monozygotic twins show concordance for FAS, compared with 64% of dizygotic twins (6). Strain-specific differences in ethanol sensitivity are described for most major model organisms (7, 8). Although the direct cause of such variability is unknown, in PNAS Dou et al. (9) suggest that alteration to ERK signaling and L1 function may be at play.

Dou et al. (9) demonstrate that ERK signaling is involved in ethanol inhibition of L1-mediated cell adhesion. The authors demonstrate that inhibition of ERK signaling blocks ethanol inhibition of L1 adhesion and show that blocking phosphorylation of an ERK site on L1 blocks ethanol-induced loss of adhesion. Dou et al. show that cell lines and mouse strains that are more susceptible to ethanol-induced effects have higher levels of ERK signaling. These results point directly to a potential mechanism for variability within FASD and hint at potential explanations for several of ethanol's other variable effects. However, the current studies should be extended to show that manipulation of ERK signaling in vivo results in changes in alcohol action on L1 and on expression of signs of FASD.

The L1 cell-adhesion molecule has numerous functions during neural development, and analysis of L1 across model systems points to the importance of L1 in neurite outgrowth, axon pathfinding, and fasciculation (10–13). In humans, mutation of L1 causes CRASH syndrome, an X-linked neural developmental disorder characterized by agenesis of the corpus callosum and hydrocephalus (14, 15); like FASD, CRASH phenotypes are highly variable.

FASD defects are similar to some of those observed in CRASH syndrome, leading to the hypothesis that ethanol may disrupt L1 function (16). In 1996, ethanol was first shown to be a potent inhibitor of L1-mediated adhesion in NIH/3T3 cells (16). Subsequently, ethanol was demonstrated to attenuate L1-mediated neurite outgrowth of cerebellar cells (17) and L1 was shown to be neuroprotective against ethanol-induced neural cell death (18). Ethanol alters growth cone dynamics and L1 function can restore proper response to guidance cues (19). Using photoactivatable forms of butanol and octanol, an ethanol-binding pocket was discovered between the interface of the first and forth Ig domains of L1 (20), suggesting that ethanol may alter the conformation of the extracellular domain.

Several intracellular effectors mediate L1 signaling and function, including ERK (21). Data presented by Dou et al. support the intriguing model in which an intracellular effector, ERK, modulates the severity of ethanol teratogenesis by altering presentation of the L1 extracellular domain ("inside-out signaling"), which in turn allows ethanol entry to its binding pocket (9). Thus, an intracellular phosphorylation event alters an extracellular alcohol interaction site (Fig. 1).

Interestingly, a strong genetic modifier exists on chromosome 5 in the C57BL/6J mouse strain, compared with 129/Sv mice, which associates with more severe hydrocephalus in L1 mutants (22). Although this region contains ~565 genes, it is important to determine if genes regulating ERK signaling are present in this interval, because Dou et al. show C57BL/6J mice have higher levels of ERK signaling. Depending upon the context, ethanol can elevate or attenuate ERK signaling (23), suggesting that the precise cellular context may determine whether L1 is inhibited by ethanol. It will be of great interest to determine if there are regional differences in ERK activation and L1 function following ethanol exposure in this unique model. Overall, the manuscript by Dou et al. (9) suggests that a complex interaction of genetic modifiers may regulate L1–ethanol interactions during development.

Aalthough the developmental roles of L1 have been most extensively studied, L1 also functions in the adult. In the hippocampus, regenerating fibers express L1 (24). L1 also functions in the hippocampus to modulate long-term potentiation (25) and memory

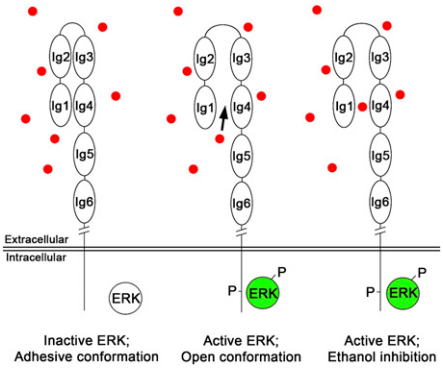

Fig. 1. ERK activation promotes ethanol-mediated inhibition of L1 adhesion. (*Left*) When ERK is inactive, L1 is in a horseshoe conformation, preventing ethanol (red dots) from reaching its binding pocket at the Ig1 and Ig4 interface. (*Center*) Activated ERK phosphorylates L1 altering the conformation of the extracellular domain allowing ethanol access to the binding pocket (arrow). (*Right*) Ethanol binding at the interface of Ig1 and Ig4 prevents L1-mediated adhesion.

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consolidation (26, 27). Chronic alcoholism results in memory and other neurological defects consistent with hippocampal impairment (28). Additionally, genes involved in MAPK signaling are elevated in mouse strains that show higher levels of voluntary drinking (29). Collectively, these findings may suggest a hypothesis in which the same pathways that promote ethanol teratogenesis also promote ethanol-mediated neural dysfunction and alcohol-seeking behaviors.

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