ABSTRACT

EFFECTS OF METHYLMERCURY ON CEREBELLAR GRANULE CELLS

OF THE TOTTERING MOUSE

By

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Methylmercury (MeHg) is an organic highly toxic form of mercury and a persistent environmental neurotoxicant. The most common manner in which humans are exposed to MeHg is by consumption of contaminated fish. Studies from cases of chronic and acute human poisonings have revealed that MeHg causes a massive loss of cerebellar granule cells (CGCs), causing the characteristic dysarthria and ataxic signs. Previous research has found that acute treatment of rat CGCs with MeHg in vitro causes a time- and concentration-dependent increase in intracellular Ca2+ ([Ca2+]i

) that is sufficient to cause CGC death. Voltage-gated Ca2+

channels (VGCCs) are believed to play a role in the mechanism of MeHg-induced cytotoxicity by possibly facilitating access for the metal to intracellular targets. Many subtypes of VGCCs are expressed in CGCs, each characterized by different pharmacological and kinetic properties. The endeavor of the present studies was to investigate the effects of MeHg on CGCs of a mouse model of human Cav2.1 (P/Q-type) channelopathy. The tottering (tg) mouse is the result of a non-lethal deleterious point mutation in the α1A pore-forming subunit of the P/Q-type Ca2+ channel. This VGCC subtype plays a crucial role in the process of neurotransmitter release in mature CGCs of humans and animal models. Data will show that, in low-K

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conditions (closely

mimicking their mature state) CGCs isolated from mice homozygous (tg/tg) and heterozygous (+/tg) for the tg mutation present a delay in the onset of MeHg-induced [Ca2+]i increase in vitro.

On the other hand, when they are acutely exposed to MeHg under depolarizing environments (mimicking their state in early development) tg/tg CGCs show increased susceptibility to cytotoxicity. Cerebellar organotypic slices from postnatal day (PND) 23-25 (after onset of ataxia) mice were also used to study the response of CGCs to MeHg taking into account the entire local cerebellar circuitry. Interestingly, +/tg cerebellar organotypic slices showed a higher percentage of CGC death than WT and tg/tg. The effect of MeHg on +/tg CGCs was partially eliminated by pretreating the slices with ω-conotoxin GVIA, an N-type VGCC antagonist. This suggests an important role of N-type VGCCs in the greater susceptibility of mature +/tg CGCs to MeHg. Of great importance is that tg-like mutations have been linked to human disorders including episodic ataxia type 2, familial hemiplegic migraine and spinocerebellar ataxia type 6. This work presents evidence of a genotype-environment interaction that could potentially identify human populations with higher risk for the neurotoxic effects of MeHg