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In vivo voltage-sensitive dye imaging in a mouse model of a human genetic epilepsy

Promotionsfach: Physiologie
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G2R43Q mice carry a point mutation in the γ-subunit of the ligand-gated $GABA_A$-receptor, the most widely distributed inhibitory receptor of the central nervous system. First discovered in humans the mutation is associated with the phenotype of childhood absence epilepsy (CAE) and febrile seizures (FS). Absence epilepsy symptoms consist of sudden loss of consciousness, arrest of normal movement, staring and stereotypical repetitive movements that last approximately 5-20 seconds, accompanied by typical 3 Hz spike-wave-discharges (SWD) on the electroencephalogram (EEG).
Remarkably, the G2R43Q-mutation introduced into the mouse, produces a very similar clinical and para-clinical phenotype as shown by previous behavioural and electroencephalographic studies. Thus the G2R43Q-mouse models the human condition well and is potentially of high clinical relevance.
All epilepsies are paroxysmal syndromes that affect entire neuronal cell populations. It is thus crucial to monitor large brain areas at high spatial and temporal resolution in order to understand the pathogenesis of these diseases. Therefore in the present study using the voltage-sensitive dye (VSD) RH 1691 and a fast imaging system (Fuji Deltaron HR1700) subthreshold neuronal responses to ramp-and-hold whisker stimulation and spontaneous activity in the barrel region of the primary somatosensory cortex were imaged in Urethane-anaesthetised genetically altered mice and controls (mean: postnatal day 48) at a frame acquisition rate of 200 Hz. Pentylenetetrazol (PTZ) -a seizure-facilitating drug- was injected intraperitoneally during the experiments while voltage-sensitive dye recorded cortical activity was compared in both states before and after the injection. A second set of experiments was conducted in adolescent mice (mean: postnatal day 20) during an age before the onset of frank epileptiform activity can be seen on EEG-recordings.
In G2R43Q-mice and controls of both age groups PTZ caused an increase of response amplitudes. The response amplitudes between G2R43Q-mice were more variable after PTZ-injection when post-PTZ responses were normalised to the pre-PTZ mean amplitude (+/d: n = 15, +/+: n = 11; Fisher's exact test: p=0.04). However, amplitude height was unaltered in G2R43Q-mice compared to control animals (t-test: p=0.67). Spontaneous activity - measured as standard deviation (SD) of VSD-fluorescence change - (+/d n = 23, +/+ n = 24) was higher on trend level in G2R43Q after PTZ injection, however, this difference was not statistically significant (Mann-Whitney-U-test: p = 0.0656). The area of lateral spread was increased after PTZ-injection in both groups, but not larger in G2R43Q-mice than in controls.
In summary, the present study provides the first *in vivo* imaging results of a recently created mouse model for human genetic absence epilepsy. In synopsis with previous *in vitro* and - yet unpublished - electrophysiological *in vivo* results in G2R43Q-mice the present data support the hypothesis of a cortical defect causing absence seizures/SWD in this mouse model. The present data are in line with the hypothesis of unimpaired fast GABAergic synaptic inhibition in G2R43Q-mice. Alternative explanation models such as impairment of tonic extrasynaptic GABA-currents are in principle compatible with the current results, but require further investigation.