KIO-300/BENAQ Modulates Epileptiform Activity in an Ex Vivo Hippocampal TLE Model

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BACKGROUND

KIO-300 (BENAQ) is an ion channel modulator currently being developed to restore vision in patients with retinal degenerative diseases. In the degenerating retina, spontaneous hyperactivity ("background noise") occurs, which reduces visual signal transmission and visual sensitivity. Prior studies demonstrated that BENAQ reduces pathological retinal hyperactivity by approximately 50%, suggesting a broader "membrane calming" effect on hyperexcitable neurons.[1,2]

Because neuronal hyperexcitability is a hallmark of epilepsy, these findings suggested that BENAQ may have therapeutic potential beyond ophthalmology. Temporal lobe epilepsy (TLE) was therefore selected as an exploratory indication to evaluate whether BENAQ could suppress seizure-associated electrophysiological activity in an ex vivo brain slice model.

Despite the availability of numerous anti-seizure drugs, approximately one-third of epilepsy patients remain inadequately controlled, highlighting the need for therapies with novel mechanisms of action.

[1] Telias M, et al. J Neurosci. 2019
[2] Tochitsky I, et al. Sci Rep. 2017

PURPOSE

To evaluate the effect of BENAQ on seizure-associated electrophysiological activity in an ex vivo mouse temporal lobe epilepsy (TLE) model and compare its activity to vehicle and the anti-epileptic reference compound phenytoin.

METHODS

Temporal lobe epilepsy was induced in C57BL/6NHsd mice via unilateral intra-amygdala kainic acid administration to generate a hyperexcitable epileptic phenotype. Brain explants were collected, sectioned into hippocampal slices, and maintained in artificial cerebrospinal fluid (ACSF) containing low Mg^{2+} to promote spontaneous epileptiform activity. Electrophysiological recordings were performed under low lux, red-light conditions. Local field potential recordings were obtained to quantify seizure-related epileptiform activity, including event frequency and signal intensity (area under the curve, AUC). Experiments evaluated BENAQ (300 µM; n=22 slices) relative to vehicle (n=22 slices), and phenytoin (100 µM; n=11 slices). Electrophysiological activity was analyzed during pre-treatment, drug exposure, and washout periods.

RESULTS

BENAQ demonstrated sustained suppression of epileptiform activity (Figure 1) in the ex vivo TLE model. In hippocampal CA1 slices, treatment with BENAQ reduced spontaneous epileptiform event frequency compared with vehicle-treated controls, with statistically significant inhibition beginning at 42 minutes following treatment (Figure 2, $p < 0.0001$). Analysis of summed epileptiform burden (AUC sum) also showed a significant reduction following BENAQ exposure (Figure 3, $p<0.0001$). Importantly, the inhibitory effect of BENAQ persisted throughout the treatment and washout periods, suggesting prolonged retention and sustained activity within neural tissue. Comparison of evoked transmission among all conditions showed that neither drug treatment, Phenytoin nor BENAQ, had a deleterious effect on electrical transmission broadly (Figure 4). Therefore, the effects of phenytoin and BENAQ can be interpreted to be specific to suppression of spontaneous epileptiform activity.

Figure 1: Example of epileptiform pre- and post-BENAQ

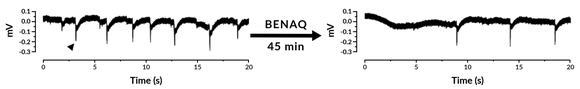


Figure 2: Effect of BENAQ (60 minutes treatment) on spontaneous epileptiform event frequency in hippocampal CA1 slices

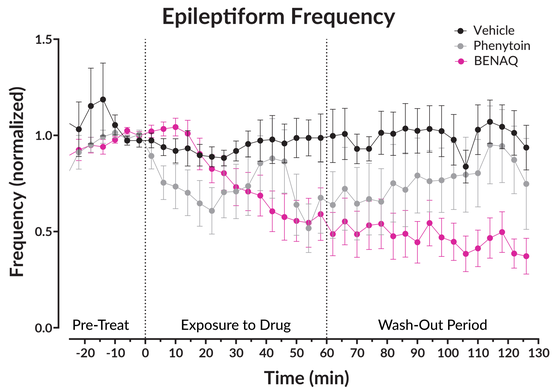


Figure 3: Effect of BENAQ (60 minutes treatment) on summed epileptiform burden (AUC sum) in hippocampal CA1 slices

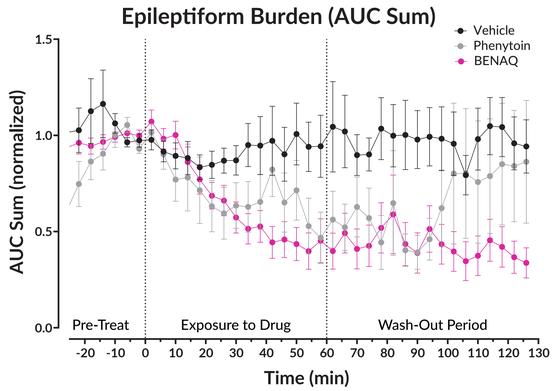
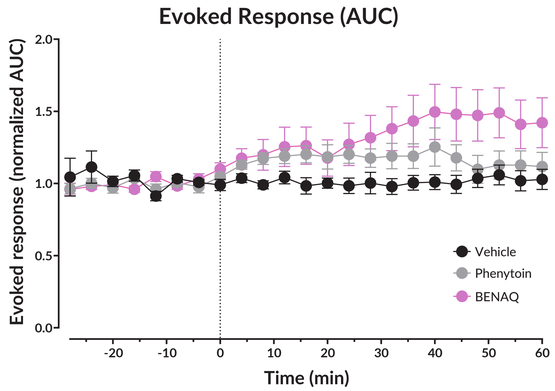


Figure 4: BENAQ does not impair evoked neural transmission



CONCLUSIONS

- BENAQ demonstrated sustained suppression of epileptiform activity in an ex vivo TLE model.
- Treatment with BENAQ reduced seizure-associated electrophysiological activity in hippocampal CA1 slices, with effects persisting throughout the washout period.
- BENAQ was not toxic to the brain slices as evoked neural activity was retained.
- These findings support further evaluation of BENAQ and related compounds in epilepsy and other hyperexcitable neurological disorders.

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