NEUROPROTECTIVE EFFICACY OF THE THIAMINE HIGH DOSE ADMINISTRATION IN CORNEA OF CHRONICALLY ALCOHOLIZED RATS

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Background and Aim. The cornea is the most densely innervated structure in the body. Corneal nerves perform sensory functions, regulates reflex tear production, blinking, and the release of trophic factors. Among many negative effects on the eye, long-term ethanol (EtOH) consumption may induce damage to corneal nerves and cause epithelial defects leading to vision impairments and possible blindness. Several mechanisms have been proposed to explain EtOH-related corneal injury, and B_1 (thiamine) hypovitaminose caused by EtOH is one of the predominant. Thus, the aim of the present study was to investigate if thiamine high dose is able to alleviate EtOH-induced corneal neurotoxicity in chronically alcoholized rats.

Materials and Methods. White albino male rats consumed 15 % (v/v) EtOH for nine months. One week before the termination of the experiment, four of EtOH-exposed rats were given thiamine (25 mg kg/b.w.). Then, corneas were isolated and lysed in RIPA-buffer by ultrasound disintegrator and centrifuged. The functional state of corneal nerves and their supportive cells was assessed by expression of their cell-specific markers (nuclear marker NeuN, neurofilament heavy subunit 200 (NF-H), and tau-protein for neurons; glial fibrillary acidic protein (GFAP) for astrocytes; and microglia/macrophage ionized calcium binding adaptor molecule 1, Iba-1) by western blot analysis. The immunodensity levels of each studied protein were normalized to actin content and then expressed as arbitrary units (a.u.).

Results and Discussion. Densitometry analysis of blots (Fig. 1) revealed that EtOH long-term exposure decreased the levels of neurospecific protein markers, NeuN and NF-H, by 3.2 and 10.8 folds, respectively, compared with control ($P<0.01$), indicating neurodegenerative changes in the cornea. Chronic EtOH consumption induced tau protein accumulation (by 7.2 folds, $P<0.01$) considered as a hallmark of tauopathy development, which impairs regeneration of corneal sensory nerves. Thiamine treatment of EtOH-exposed rats significantly restored both NeuN and NF-H levels and accelerated tau protein turnover in the cornea.

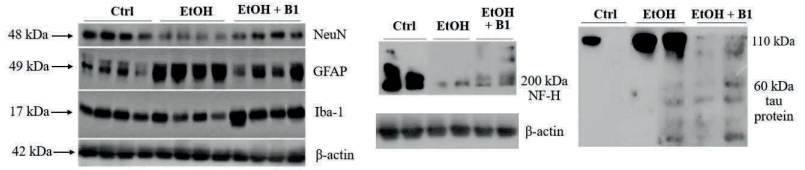

Fig. 1. Thiamine (vitamin B_1) alleviated pathological changes in expression of neurospecific proteins in the cornea of rats chronically exposed to ethyl alcohol (representative blotograms)

We showed that EtOH up-regulated corneal GFAP (by 4.3 folds compared with control, $P<0.01$) and slightly, but statistically significantly, down-regulated Iba-1 levels (by 1.5 folds, $P<0.05$) that might be considered as hallmarks of astroglia activation and macrophage function suppression during EtOH consumption. Vitamin B_1 high dose administration diminished GFAP level in the cornea of alcoholized animals indicating restriction of reactive astrogliosys. In contrast, B_1 treatment induced elevation of Iba-1 level in the cornea of EtOH-affected rats (by 2.8 folds compared with EtOH group, $P<0.05$), suggesting activation of Iba-1-positive macrophage population that is required for removal cellular debris in the injured corneal tissue.

Conclusions. Our data indicate that thiamine high dose administration could be beneficial for minimizing the neurological consequences of long-term ethyl alcohol abuse in the cornea and may provide cost-effective and productive route to ocular disease intervention.