ROLE OF NT3/TRKC IN THE REGULATION OF FEAR

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Background The most common mental disorders in EU are anxiety disorders, with an estimate of 25 million people being affected. Anxiety disorders affect all aspects of patients' life, posing a huge economic burden for society. Neurotrophins are essential for the development and function of the nervous system and have been associated with mental disease. Our own research shows that neurotrophin 3 (NT3) and its receptor TrkC regulate pathological fear[1] – a shared feature of anxiety disorders. Previous experiments pointed also to a putative role of NT3-TrkC in the regulation of contextual fear conditioning (CFC) and extinction (CFExt)[2], paradigms translationally relevant to disease.

Aims This project aims to investigate NT3-TrkC role in the formation and extinction of fear and elucidate on the underlying molecular and cellular mechanisms.

Methods C57Bl/6J animals were trained in the CFC and CFExt paradigms and sacrificed at specific timepoints of fear processing. The hippocampus, prefrontal cortex and amygdala were dissected to determine TrkC expression and activation levels by Western blot, or to isolate synaptoneurosomes to determine the synaptic expression of TrkC. Moreover, the effects of NT3 stimulation on the synaptic expression of NMDA and AMPA receptor subunits in cultured hippocampal neurons were characterized by immunocytochemistry.

Preliminary results Animals were categorized in *extinction-success* (>30% reduction in freezing levels) and *extinction-failure* (<30%) groups. Interestingly, a correlation was found between anxiety in the elevated plus maze and extinction performance, i.e. animals that fail to extinguish fear also show higher levels of anxiety. Western blot results showed a spatio-temporal complex pattern of TrkC (in)activation. Of relevance, after fear extinction acquisition, increased activation of TrkC was found in the amygdala in the *success* group, indicating a possible involvement of TrkC signalling in extinction consolidation. Moreover, immunolabeling of synaptoneurosomes showed that extinction acquisition is associated with increased TrkC recruitment in amygdala synapses. Finally, incubation of cultured hippocampal neurons with NT3 for 10 min increased the synaptic expression of GluA1 but decreased GluA2, while 30 min incubation increased the synaptic expression of both GluN2A and GluN2B. Glutamate receptors regulation may represent a mechanism for the role of NT3-TrkC signalling in conditioned fear and extinction.

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