

Mood disorders (e.g., depression) and anxiety are among the most common mental disorders identified in populations throughout the world. They affect all age groups, from children to adults. Despite many studies and many existing theories the etiology of these disorders is not well understood.

Early-life stress appears to be a particularly important risk factor for psychiatric disorders. Clinical and epidemiological studies proved that persons who experienced an adverse experience in the initial period of their life have increased vulnerability to mood disorders, anxiety disorders and are more prone to addiction. The most negative impact on development of individual has a dysfunctional family. According to a study published by the World Health Organization WHO more than 50% cases diagnosed in children, more than 30% cases diagnosed during adolescence and 13% of adult cases of mood disorders is associated with adverse experiences in early life. These results show that early-life stress leads not only to increased risk of mental illness, but also greatly accelerates its outcome. It should also be noted that not all individuals experiencing the early-life stress develop previously described mental illness. Research that use animal models indeed indicate that in some individuals early-life stress leads to adaptation and better coping with unpleasant experiences in later life. Previously described effects of early stress are reflected in changes in brain structures. Prefrontal cortex (mPFC) is one of the brain structures whose malfunction is commonly associated with mood and anxiety disorders. It is located in the frontal part of forebrain. It is responsible for higher cognitive functions, decision making, risk assessment and integration of information coming from other parts of the brain. The mPFC is one of the structures characterized by prolonged developmental trajectory. It means that it reaches maturity later than other parts of the nervous system. Long development of final function of this structure entails its high vulnerability and susceptibility to changes caused by environmental factors in the postnatal period. Rapid development and modifications within this structure takes place in adolescence. It is a time associated with the personality changes. Adolescents are more impulsive, willing to take risk, more susceptible to environmental influences and also more rapidly respond to stress.

In our previous studies using an animal model of early-life stress, i.e., repeated maternal separations of rat pups for a period of 3 hours on postnatal days from 1 to 14, we have shown the effect of early-life experiences on the structural and functional changes in the mPFC. The results have shown a reduction in density of nervous cells extensions (dendritic spines) and impairment of long-term potentiation (LTP) a process underlying learning and memory formation which is a form of adaptation to the environment i.e., synaptic plasticity. Moreover animals subjected to the separation procedure were characterized by increased anxiety and impaired fear memory. Despite the obtained results and data from other scientific reports it is still little known about the functioning of the mPFC during adolescence.

Presented facts together with results of our previous research prompt us to propose a project in which we plan to investigate influence of early-life stress on changes in the mPFC and corpus callosum (CC). The CC is the main tract of white matter that is responsible for communication between hemispheres. In our research we will use animal model that was previously described. We are going to measure the volume of mPFC and CC, estimate the numbers of basic cells that build nervous tissue i.e. neurons and glial cells in animals reared in conventional facility rearing and those which undergone the maternal separation procedure. We would like to focus especially on glial cells as few data report that its role is not limited to supportive functions, synapse elimination and myelination but they are also involved in processes underlying the functional synaptic plasticity. Moreover changes in glial cells activity are also observed in many psychopathologies, like depression or neurodegenerative disorders. Additionally, in this part of the project we plan to investigate serum expression of protein markers, which are related to glial cells activity. In the next step we would like to examine influence of acute stress of different intensities on plasticity and glial cells functioning in the mPFC. The aim of this stage is to get closer to an answer for a question if early-life stress causes an adaptation in later life and allows to better cope with similar situations in later-life or adverse experiences cumulate causing more rapid reaction and vulnerability. To mimic physiological stress, we will use acute corticosterone treatment. Corticosterone is an animal equivalent of cortisol. It is a hormone produced by adrenal glands cortex. Corticosterone concentration in blood increases rapidly in reaction to stressful stimuli. We plan to pay particular attention on that how the early-life stress together with corticosterone treatment modulate the LTP and basal synaptic transmission. Experiments are also planned to determine what types of cell are activated and also how early-life stress together with corticosterone treatment influence molecular processes underlying functional synaptic plasticity. We would also like to investigate mRNA and proteins expression of synaptic plasticity markers. In order to more comprehensively examine influence of early-life adversity on adolescents it is also planned to investigate the level of corticosterone binding proteins i.e., albumins and corticosterone binding globulin (CBG). These proteins are responsible for corticosterone transport and they play important role in regulation of corticosterone concentration in blood. We decided to do that because recent studies have reported that CBG determine behavioral response to stressful situation. In this part of the project behavioral tests will be also used, during those tests we will examine anxiety and fear memory performance after acute corticosterone treatment. Those research will allow to correlate functional changes in neural network with behavioral outcome.

To achieve presented goals we are going to use the following research methods. In order to mark specific types of cells we will use immunohistochemistry, whereas expression of different proteins will be assessed using Western blot. In both of those methods proteins are marked with specific antibodies. In order to measure gene expression level we are going to use quantitative PCR. Basal synaptic transmission and LTP will be measured using electrophysiology. Behavioral tests aimed to assess the level of anxiety will be conducted using light/dark box test, whereas fear-memory will be measured in classic Pavlovian fear conditioning.

Summing up the briefly presented assumptions and research plan, this project aims to find structural and biochemical markers of early-life stress related psychopathologies and explain processes and mechanisms which underlie them. Modern neuroimaging techniques together with this knowledge may ease early diagnosis and risk assessment of mood and anxiety disorders in individuals that were subjected to early-life stress. Equally important problem that will be investigated during this project will be an attempt to define what determines the final outcome of early-life stress and why its effects can vary between individuals. It cannot be also ignored that results of the project will be important for better understanding how particular mechanisms combine and act together to determine individual behavior in face of different situations and how this behavior is influenced by environmental factors and earlier experiences. Proposed research may also contribute to an increase in social awareness of children's rights protection through showing irreversible effects of stress experienced during childhood.