1. Research project objectives/ Research hypothesis

The pathophysiology of autism spectrum disorders (ASD) is still unexplained. Part of the research underlines the significant role of genetic factors in the etiology of this brain disorder. Previous work indicates a number of genes that disturbed expression may result in neurodevelopmental dysfunctions leading to the development of ASD symptoms. According to the *Developmental Origins of Health and Disease* concept, environmental factors such as maternal diet during pregnancy and lactation, determine the susceptibility to diseases that manifest themselves later in life. In recent years, there has been a rapid increase in the number of environmental factors studied that may be directly related to the development of autism, including the role of maternal diet components.

A balanced maternal diet is necessary for proper fetal brain development and limiting nutrients or consuming unhealthy foods during intrauterine and early childhood life is associated with a strong increase in the risk of neurological diseases as well as other neurodevelopmental disorders. One of the mechanisms that can explain changes in the expression induced by diet genes are epigenetic changes by methylation of cytosine residues in CpG dinucleotides, important - among others - in neuronal development.

The aim of this project is to assess the impact of a modified diet consumed during pregnancy and lactation by the mother, that may change the expression of genes - identified with the risk of developing complex and multifactorial ASD - in the prefrontal cortex (PFC) of both sexes in the offspring. In addition, linking environmental factors with genetic changes, by determining the degree of methylation of CpG islands, promoter sections of selected genes may allow to explain the molecular mechanisms through which pathology develops. In addition, a detailed description of this type of effects will allow a better understanding of the role of maternal diet in the early stages of offspring as an important factor that can generate predisposition to an increased incidence of ASD.

2. Research project methodology

Our studies indicate that offspring from mothers which were fed modified diet shows a phenotype characteristic of autistic disorders, including anxiety-like and cognitive disturbances. The project includes multilevel studies including the assessment of the impact of modified types of maternal diet on male and female offspring, including diet: rich in carbohydrates, high fat, mixed and standard, consumed during pregnancy and lactation on differences in gene expression in the prefrontal cortex, which are strongly associated with ASD. Biochemical and molecular analysis of this project include both gene analysis and the proteins they encode. Screening analysis of the expression of larger genes will be carried out using expression matrix cards. Next, the mRNA levels of the genes selected on its basis will be validated using the RT-qPCR method. In order to explain the potential mechanism that can induce changes in expression, an analysis of methylation of CpG islands of promoter sections will be performed. Using the immunoenzymatic ELISA assay and the Western Blot, the level of proteins encoded by genes characterized by reduced or overexpressed will also be assessed to verify functional changes.

3. Expected impact of the research project on the development of science

ASD is still a poorly understood, severe neurodevelopmental brain disorder, often leading to the exclusion of a patient from normal life in society. The complexity of the basis of this disease has not yet allowed to fully understand its neurobiological mechanisms and factors that may induce the development of ASD. Maternal nutrition as a necessary element for fetal brain development can be an important element determining the risk of developing symptoms of ASD. Planned research in the project will be the first study focusing so strongly on the assessment of the impact of the modified maternal diet on the risk of genetic modification predisposing the offspring to ASD. An in-depth understanding of the role of factors such as the maternal diet can contribute to a better understanding of the mechanisms of this complex disease and provide a basis for seeking new prevention and treatment options for ASD. Therefore, the results of this project may have an impact in the future on reducing the medical, social and economic damages that the maternal diet may have by affecting the offspring's health.