Introduction

Idiopathic scoliosis (IS) is a three-dimensional spine deformation, which occurs in otherwise healthy children and adolescents. The prevalence is 2-3% in adolescent population. The patients do not reveal congenital vertebrae malformations, neuro-muscular or other diseases. Idiopathic scoliosis occur in varies clinical forms, differing in magnitude, number and localization of the curvatures. IS may reveal in two major forms: 1) small curvature, stable through the life that does not need to be treated, or 2) progressive form which has tendency to rapid curve progression to big deformations that has detraining influence on the body function, shortens the life length expectation and decreasing quality of life. The differentiation between these two forms is at the early stage of the very difficult based on currently used examinations. IS progress much more often in girls than in boys, this disequilibrium increases with the curvature severity and for the curvature angle above 30° is 7:1. Thus the factors associated with puberty seems to play important role in etiopathogenesis IS.

It is multifactorial disease with genetic and environmental background. Polimorphisms of many genes were described to be associated with IS. Among them estrogen receptors were evaluated. Although the there were not direct influence on occurrence of IS, they may have function modifying disease course. Another gene associated with IS occurrence is ladybird homeobox 1. The association of polymorphisms of LBX1 were found in GWAS and replicated in many studies. The *LBX1* gene encodes a regulatory protein, which expression determines the properties of the migration of the myogenic cells. The gene is essential for the recognition of signals, which give direction and maintain their migratory potential. Among evaluated genetic associations of varies genes polymorphism with IS. The polymorphisms that revealed association with IS are more common in IS patients than in control group, however, they are also present in healthy people without IS. That indicates that there are some other factors that are needed to develop IS

Nowadays there are publications suggesting that the modifying agents in IS may be epigenetic factors. epigenetic factors are not associated with DNA sequence, however, they may influence the gene expression. Despite well described hypotheses suggesting association of the epigenetic factors with IS, to our knowledge there is no single study analyzing this topic.

Aim

The aim of this projects is an analysis of the influence of the epigenetic factors, namely the global DNA methylation, and the local methylation of CpG islands of estrogen receptors type 1 and type 2 genes (*ESR1, ESR2*) and ladybird homeobox 1 gene (*LBX1*) on predisposition to IS and clinical form of IS.

Method

The study consists of two parts: clinical (clinical and radiological examination and samples harvesting) and molecular (global DNA methylation analysis, *ESR1, ESR2* and *LBX1* methylation and expression analysis).The study group will consist of female patients treated operatively due to IS in Department of Spine Disorders and Pediatric Orthopedics University of Medical Sciences in Poznan. The control group will consist of female patients after spine surgery performed for different reasons but not IS. After informed consent obtaining from each patient, the samples of peripheral blood as well as the samples of paraspinal muscles from the superficial and the deep layer will be collected. DNA will be extracted from blood samples while DNA, RNA and proteins will be extracted from muscle tissue. The molecular analysis will be based on global methylation of the genome evaluation in blood and muscles samples. The local methylation level of CpG islands *ESR1, ESR2* and *LBX1* will be assessed with pyrosequencing method. The expression of *ESR1, ESR2* and *LBX1* will be analyzed to determine the influence of methylation on the expression profile. The global methylation profile in patients with IS will be compared between results from blood and muscles and between different layers of the muscles samples. The global methylation will be compared between the study and control group and within IS patient's subgroups. The correlation between methylation level in the blood and muscles samples will be performed. The level of *ESR1, ESR2* and *LBX1* methylation will be compared between the study and control group and within IS patient's subgroups. At the last step influence of the local methylation of *ESR1, ESR2* and *LBX1* on the of expression in the muscles will be performed.

Reasons of topic evaluation

Taking into consideration the prevalence of IS in population, it is both medical and social problem. The deficiency of sufficient knowledge concerning IS background and its pathogenesis is the reason that the treatment of this disease is symptomatic (spine surgery, corrective braces), instead of causal approach to the disease.

Analysis of the presented hypothesis explain unparsed until now problem and will have significant influence on knowledge concerning etiopathogenesis of IS. In case of positive confirmation of our hypothesis, not only some part of IS etiology will be explained, but also possibility to pathogenetic path analysis will be possible to discover and early treatment endorsed.