DNA methylation status and expression of pluripotency related genes in testicular development of rat

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SALL4 and LIN28A/B are pluripotency related markers, being highly expressed in testis in undifferentiated spermatogonia respectively, where they play an essential role in maintaining their pluripotent and self-renewal properties. However, data about their DNA methylation dynamics during testicular development are limited. The aim of this study was to analyze and compare expression dynamics with DNA methylation status of SALL4 and LIN28A/B in different stages of early fetal and neonatal testicular development of rat. DNA and RNA were isolated from the fresh fetal and neonatal samples of rat testis taken successively from GD20 to PND5,5. qPCR method was applied to determine expression status of SALL4 as well as LIN28A/B while the pyrosequencing method was used to determine the level of CpG methylation in promotor regions of mentioned genes. The results had demonstrated that SALL4 and LIN28A/B were expressed in all examined testicular developmental stages with significantly higher mRNA expression in the fetal compared to the early neonatal stages of testicular development. Furthermore, promoter regions of both genes were highly hypomethylated, however, without a difference in the level of methylation between the individual developmental stages. We conclude that the expression and related function of spermatogonial –related genes could be controlled at the posttranscriptional level, which is a much quicker way to activate/deactivate genes than the DNA methylation mechanism. This study was supported by the Scientific Center of Excellence for Reproductive and Regenerative Medicine, Croatia, and through the European Regional Development Fund, under grant agreement KK.01.1.1.01.0008. „Reproductive and Regenerative Medicine-Exploring New Platforms and Potentials”.