

P0454

The Regulation of the Testicular Rhythm Coordinated with Circadian Clock Genes

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Circadian rhythms, which measure time about 24 hours, are generated by one of the most ubiquitous and well investigated timing system. More recently, circadian clock gene expression has been reported in various peripheral tissues. If a circadian clock is functioning in the testis, expression of clock genes should be observed in this tissue. To resolve this issue, we examined the expression of circadian clock genes in the testis.

Expression of mRNA of *Period1 (Per1)*, *Period2 (Per2)*, *Period3 (Per3)*, *Cryptochrome1 (Cry1)*, *Cryptochrome2 (Cry2)*, *Clock*, *Bmal1*, *Timeless (Tim)*, *Prokineticin1* and *Prokineticin2 receptor (Prok1r, Prok2r)* genes in mouse testis was explored by semiquantitative reverse transcription polymerase chain reaction (RT-PCR) ; 1) According to the circadian time(CT 2, 6, 10, 14, 18, 22, constant dark(DD) and light dark(LD)). 2) According to the developmental stage(post partum 1day, 7day, 10day, 21day, 35day) Immunohistochemistry using PER1 was analyzed.

Expression of the circadian clock genes *Per2*, *Per3*, *Bmal1*, *Clock*, *Cry1* and *Cry2* was constant at all times, except for *Per1* and *Tim* in mouse testis. Immunohistochemical localization of PER1 was revealed at all times of the day. Expression of PER1 was a restricted only in cells at the spermatogonia and the condensing spermatids. According to the developmental stages, clock genes were highly expressed at ppd 7 and 10. Receptors for *Prok2* were also highly expressed between ppd 1 and 7 and then decreased gradually. *Per1* and PER1 expression were increased in ppd7, decreased in ppd 10 and 21 and increased again in ppd 35.

These studies indicate that the clock gene expression in testis presented different expression compared with other tissues, like the liver. But it could be observed that the clock genes expression was related in gonadal development and germ cell differentiation in mice. More advanced studies about circadian genes function is required to conform the regulatory mechanisms of circadian genes in spermatogenesis.

Key words: *Testis*, *Circadian*, *Clock genes*, *RT-PCR*, *Immunohistochemistry*