



Treatment of Immature Mice with Cyclophosphamide Decreased the Amount of Spermatogenic Cells and Somatic Cells that Produce Growth Factors in the Testis

Ronnie Solomon¹, Ali Abu Madigim¹, Maram Abofoul-Azab¹, Eitan Lunenfeld², Mahmoud Huleihel¹

¹The Shraga Segal Dept. Microbiology, Immunology and Genetics, and The Center of Advanced Research and Education in Reproduction (CARER), Faculty of Health Sciences, Ben-Gurion University of the Negev, Beer-Sheva, Israel

²Dep. OB/GYN, The Center of Advanced Research and Education in Reproduction (CARER), Soroka Medical Center, and Faculty of Health Sciences, Ben Gurion University of the Negev, Beer Sheva, Israel

Introduction: The proliferation and differentiation of spermatogonial stem cells to sperm are under the regulation of testicular somatic cells such as Sertoli, peritubular and Leydig cells. Cyclophosphamide (CP) is an alkylating agents' chemotherapy that affects proliferating cells, including spermatogonial cells in the testis.

Aims: To evaluate the effect of CP on the amount of spermatogonial cells and on the amount of testicular somatic cells of immature mice.

Materials and methods: Seven days-old mice were divided into: 1) CP group that were injected intraperitoneal (i.p) with 100 mg/kg CP once a week during 3 weeks. 2) Mice of the control group (CT) were injected with saline. Mice were sacrificed 10 days after the last injection. Cells were enzymatically isolated from the seminiferous tubules, counted and stained for markers specific for spermatogonial (premeiotic), meiotic and post-meiotic cells by immunofluorescence staining (IF) using specific antibodies. Cells were also examined for growth factor expression and somatic cells by double IF staining.

Results: Our results show a reduction in the total amount of tubular cells, spermatogonial cells, meiotic and post-meiotic cells in CP-treated mice compared to control. In addition, a significant reduction was observed in the number of tubular cells mainly Sertoli and peritubular cells.

Conclusions: This is the first study that shows a direct effect of CP on the number of somatic cells that produce growth factors, and also on the different types of spermatogonial cells in immature mice. Thus, the impairment of spermatogenesis following CP treatment could be related to direct effect on both spermatogonial and somatic cells.