

C-kit Expressing Male Germ Cells were Highly Sensitive to Busulfan Treatment and Apoptosis of Male Germ Cells Induced by Busulfan Treatment was not Caused by Fas/FasL or p53

옥도원, 이미숙, 권득남, 김진희

Dept. of Dairy Science, Division of Applied Life Science,
Gyeongsang National University, Chinju, Gyeongnam, 660-701, Korea

Male germ cell apoptosis has been extensively explored in rodent. In contrast, very little is known about their susceptibility to apoptosis stimuli of developing germ cell stages at the time when germ cell depletion after busulfan treatment occurs. Furthermore, it is still unanswered how spermatogonial stem cells are resistant to busulfan treatment. Spontaneous apoptosis of germ cells was observed in the testis of adult mice and experimentally induced busulfan treated mice increased this apoptosis to such an extent that there was a decrease in the weight of the testis. One week after busulfan treatment, TUNEL staining resulted in selective degeneration of spermatogonia and some of early meiosis spermatocytes. RT-PCR results further validated the findings of TUNEL-stained apoptotic cells. The percentage of apoptotic-positive tubules and apoptotic cell index increased time dependently: an immediate effect on type A spermatogonia at 1 week after treatment and in another week that followed, secondary effect on haploid cells. RT-PCR results by using spermatogonia-specific biomarker showed that c-kit and Stra 8 expression was reduced, but that Gli 1 expression was constant, indicating the initiation of primary apoptosis of type 2 spermatogonia. Downregulation of FasL at 4 weeks after injection of busulfan contributed to the initiation of secondary apoptosis of haploid cells. Expression of FasL was inhibited while expression of Fas increased after the 2-busulfan treatment and remained at levels about two times of the control. Our results indicate that busulfan resulted in apoptotic death of testicular germ cells and that this process occurred independently the Bcl-2 family genes and the Fas signaling system.

Key words) *apoptosis, busulfan, male germ cells*