

Ruksinee Khammanit 2007: Effects of Serum Starvation, Cell Confluency and Chemical Inhibitors on Cell Cycle of Canine Dermal Fibroblast. Master of Science (Veterinary Anatomy), Major Field: Veterinary Anatomy, Department of Anatomy. Thesis Advisor: Assistant Professor Sirirak Chantakru, Ph.D. 79 pages.

The cell cycle stage of donor cells and the method of cell cycle synchronization are important factors influencing the success of somatic cell nuclear transfer. The objective of this study was to examine the cell cycle characteristics of canine dermal fibroblasts cultured under various conditions: serum starvation, culture to confluence and treated with chemical inhibitors, roscovitine, aphidicolin and colchicine. Canine dermal fibroblasts were isolated from skin samples obtained from the abdominal region of bitches during routine ovariohysterectomy. Cells were cultured and used for this experiment between passages 2 and 6 of culture. By short periods of 24, 48 and 72 h of serum starvation significantly increased ($P < 0.05$) the percentages of cells at the G0/G1 phase to 88.4 ± 1.3 , 90.9 ± 1.4 and 90.3 ± 2.2 , respectively, which was similar to culture to confluency ($91.7 \pm 3.5\%$). Treatment with different concentrations of roscovitine did not increase the proportion of G0/G1 cells, on the other hand, it significantly increased the percentage of cells that underwent apoptosis at concentrations of 30 (10.9 ± 10.9) and 45 μM (16.2 ± 12.4). Using aphidicolin led to increase percentages of cells at the G1/S transition in a dose-dependent manner without increasing of apoptosis. Colchicine at a concentration of 0.1 $\mu\text{g/ml}$ significantly increased ($P < 0.05$) the proportion of cells at the G2/M phase (38.5%), on the other hand, it significantly decreased the proportions of G0/G1 cells (51.4%). However higher concentrations of colcemid above 0.1 $\mu\text{g/ml}$ did not increase the percentage of G2/M phase cells. These results indicate that canine dermal fibroblasts can be effectively synchronized at various stages of the cell cycle, which could have benefits for somatic cell nuclear transfer in this species.

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