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Anti-tumor Effects of Canine and Human Adipose Tissue-derived Mesenchymal Stem Cells in a Xenograft Mouse Model for Melanoma

Jin-ok Ahn[1], Ye-rin Coh[1], Hee-woo Lee[1], Il-seob Shin[2], Sung-keun Kang[2], Hwa-young Youn[*1]

[1]Department of Veterinary Internal Medicine and Research Institute for Veterinary Medicine, College of Veterinary Medicine, Seoul National University, Seoul, Republic of Korea; [2]Stem Cell Research Center, K-STEMCELL Co. Ltd., Seoul, Republic of Korea

Introduction: Mesenchymal stem cells (MSCs) have been identified as promising therapeutic tools in cancer treatment since they possess a powerful capacity for tumor-directed migration and incorporation, highlighting their potential as an optimal vehicle for delivering anticancer agents. The first part of the study was performed to investigate the anti-tumor effect of human adipose tissue-derived mesenchymal stem cells (AT-MSCs) on human melanoma. The second part of the study was designed to test the hypothesis that the stem cell-based gene therapy combined with low-dose cisplatin would improve therapeutic efficacy against canine melanoma.

Materials and Methods: The inhibitory effect of canine and human AT-MSCs on the growth of melanoma cells was evaluated using a cell viability assay. Cell-cycle arrest and apoptosis in melanoma cells were investigated by flow cytometry and western blot analysis. To evaluated the in vivo anti-tumor effect of AT-MSCs, CM-DiI-labeled AT-MSCs were circumtumorally injected in tumor-bearing athymic mice and tumor size was measured.

Results: The results showed that AT-MSC-CM significantly inhibited the viability of A375SM and A375P cells. AT-MSC-CM induced G0/G1 cell cycle arrest and apoptosis in A375SM and A375P melanoma cells. Western blot assays showed that cyclin D1 levels were decreased and caspase-3, caspase-7 and PARP protein levels were increased in melanoma cells cultured with AT-MSC-CM. Treatment with AT-MSCs suppressed tumor growth in the mice and fluorescence analysis revealed that AT-MSCs migrated efficiently to the tumor tissues. In the second part of the study, the IFN-β transduced canine AT-MSCs (cAT-MSC-IFN-β) inhibited the growth of LMeC canine melanoma cells in vitro in direct as well as indirect co-culture systems. Flow cytometric cell cycle analysis showed that the proportion G0/G1 phase LMeC cells co-cultured with cAT-MSC-IFN-β was higher than that of the controls. G1 arrest occurred concurrently with a reduction in the percentage of S phase cells. In animal experiments using BALB/c nude mouse xenograft model which was established by injecting LMeC cells subcutaneously, the combined treatment of cAT-MSC-IFN-β and low-dose cisplatin significantly reduced tumor volume compared to the control and single agent treatment groups. Fluorescent microscopic analysis of the tumor section provided evidence for homing of cAT-MSC-IFN-β to the tumor site and TUNEL (terminal deoxynucleotidyl transferase-mediated nick-end labeling) assay showed that the combination treatment of cAT-MSC-IFN-β and low-dose cisplatin induced high levels of cell apoptosis.

Conclusions: Cell therapy using AT-MSCs demonstrated an anti-tumor effect on canine and human melanomas, suggesting a possible therapeutic option for this type of cancer. These findings may provide valuable information warranting further explorations of the application of the combined therapy for various tumors including malignant melanoma.

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Diversity of the Gastric Bacterial Microbiome in Thoroughbred Racehorses Having Gastric Ulcer

Hee-Jin Dong, Hungwui Ho, Hyeshin Hwang, Woohyun Kim, Yongbaek Kim, Janet Han, Inhyung Lee, Seongbeom Cho[*]

BK21 PLUS Program for Creative Veterinary Science Research, Research Institute for Veterinary Science and College of Veterinary Medicine, Seoul National University, Seoul 151-742, S. Korea

Introduction: Clear differences in the fecal microbial communities between healthy and unhealthy horses were observed in several studies, suggesting that microbiome imbalance may contribute to disease. For racehorse, equine gastric ulcer syndrome (EGUS) is one of the most frequently reported diseases, but information regarding equine gastric microbial communities is limited. In this study, to evaluate the microbiological impact on EGUS, the presence of *Helicobacter* spp. and the microbial diversity were investigated in Thoroughbred racehorse in Korea.

Materials and Methods: A gastroscopy was performed on 52 Thoroughbred racehorses within 2 days after racing at the Korean Racing Authority during August 2013 to October 2013 and the severity of gastric ulcer were scored according to the standard grading system. Horse information was obtained regarding the animal age, gender, and management factors including dietary patterns (type, frequency, and amount of feed and water), type of bedding, and past records of drug administration. Two biopsy samples were obtained to identify the presence of *Helicobacter* spp., including *H. pylori* and *H. equorum*, and to characterize the gastric microbial community using next-generation sequencing.

Results: A total of 52 gastric biopsies and 52 fecal samples were tested for the presence of *Helicobacter* species, however culture and PCR yielded negative results. The microbial communities of 11 racehorses were characterized. *Firmicutes* was the most predominant phylum in horses regardless of the severity of the gastric ulcer, but the proportions of other major phyla varied markedly with the severity of the gastric ulcer; a higher proportion of *Actinobacteria* was present in horses with mild gastric ulcers and a higher proportion of *Proteobacteria* was present in horses with moderate to severe gastric ulcers. Two clusters were generated by Unweighted Pair Group Method