5) An experimental study of the microsurgery using SHRSP. Analysis on the endothelial cell morphology of the femoral artery. Noritaka Isogai, Nozomu Kurozumi, and Hiroshi Kamiishi. Department of Plastic and Reconstructive Surgery, Kinki University Hospital, Osaka-fu 589.

INTRODUCTION

Our previous experiments of the microvascular surgery using SHRSP demonstrated an accelerated wound healing at the site of microvascular anastomosis by the scanning electron microscope observation. To throw some lights on this endothelial recovery process following microsurgical repair at the site of microvascular anastomosis, we have performed an study on the endothelial cell morphology between normotensive rat(WKY) and hypertensive rat(SHRSP) by introducing a technique of En face silver staining method.

METHODS

The animals used were 4 week old SHRSP and 12 week old SHRSP. The WKY served as a control population. The anastomosed site was histologically evaluated for the morphological study of the endothelial cells by using a modification of the method described by Poole et al(1958). After saline infusion arteries were cut open along the longitudinal axis and pinned to the wax block; the intima uppermost. Blocks were successively transferred in the dark at room temperature to the following solutions: 650 mM formaldehyde(5 min), 15 mM AgNO_3 (2 min), and 139 mM CoBr_3 with 102 mM NH_4Br (2 min). Wet preparations were directly observed en face using a light microscope at x200 magnification. Images were then directly analyzed by using the Computer Graphic Analyzer System(CGAS, TVIP 2100, Avionics Co., Ltd., Japan), whose digital analyzer measured each size of the endothelial cells and calculated the mean and the standard deviation. These data were statistically checked by using the t-test to compare the difference between values.

RESULTS

1. There was no significant difference on the shape of endothelial cells between 4 week old SHRSP and the control, however, 12 week old SHRSP showed small and spindle endothelial cell shape with irregular cellular arrangement.
2. CGAS objectively quantified the size difference of the endothelial cells stained by En face silver staining method. Average cell size of the 4 week old SHRSP was measured $310 \mu\text{m}^2$, while it was measured $333 \mu\text{m}^2$ in the control. There was no statistical significance among them. 12 week old SHRSP demonstrated an average endothelial cell size of $279 \mu\text{m}^2$, while $356 \mu\text{m}^2$ was obtained in the control. Significance of this change was $p < 0.001$, so that the average endothelial cell size in 12 week old SHRSP was comparatively smaller than the one in the control.
3. The distribution of the measured endothelial cells in each group was evaluated. A regular distribution pattern with one peak was obtained in the 4 week old SHRSP, which was quite similar to the one in the control, even though 12 week SHRSP demonstrated an endothelial cell distribution with 2 peaks at $50\text{--}100 \mu\text{m}^2$ and $250\text{--}300 \mu\text{m}^2$ cell size, respectively.

DISCUSSION

It still remains unclear how wound healing process takes place at the site of microvascular anastomosis site, especially in the diseased vessels such as the one with hypertension or arterial sclerosis. We focused our attention on the endothelial cell morphology and cell size to evaluate the basic difference between SHRSP and the control(WKY), which may lead to an answer for the early establishment of the endothelial cell repairing at the anastomosis site in the SHRSP group. A comparison between the rat wound healing and the human healing in the state of hypertension will make these findings clinically important.