

SPECTROCOLORIMETRIC EVALUATION OF REPAIRED ARTICULAR CARTILAGE IN RABBITS

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Introduction: In clinical practice, repaired cartilage is often assessed by visual inspection and probing using arthroscopy. Through an arthroscopic monitor, human eyes can differentiate color changes in repaired cartilage compared with surrounding intact cartilage, but cannot quantify these color changes. Therefore, there is a need for an objective method to evaluate the repaired tissue itself and provide a prognosis after treatment. We investigated the use of a commercial spectrophotometer and the application of two color models ($L^* a^* b^*$ colorimetric system and spectral reflectance distribution) to describe and quantify articular cartilage. In the present study, we measured the colors of intact and repaired cartilage after a microfracture using a spectrophotometer and evaluated the obtained results in comparison with the results of histological, histomorphological and biochemical findings.

Materials and Methods: Nineteen adult rabbits were used. The rabbits were anesthetized and an anteromedial arthrotomy was performed in left knee. The patella was dislocated laterally and the articular cartilage was resected with a chisel to form a 5 mm-diameter defect down to the subchondral bone. The defects were washed with saline and dried with a swab to remove any debris. Subsequently, a microfracture technique was used in the defect. An awl was then used to make multiple holes in the defect, and the wound was then closed in layers with 2-0 vicryl sutures. The right knee was left without treatment as a control (group C). The rabbits were sacrificed at 2 (group M-2; n=5), 4 (group M-4; n=7) and 12 (group M-12; n=7) weeks after the operation. The cartilage samples were used for spectrophotometric assessments.

Spectrophotometric measurements

Articular cartilage evaluation was performed using a spectrophotometer (X-Rite SP64; X-Rite K.K., Tokyo, Japan). The X-Rite SP64 was positioned with minimal pressure perpendicular to the cartilage defect area or the intact cartilage area as a control. Three consecutive measurements of the L^* , a^* and b^* values and the spectral reflectance ratio per site were averaged for each cartilage measurement.

After spectrophotometric evaluation, specimens were used for histological, histomorphological and biochemical assessments.

Results: Spectrophotometric measurements

The L^* value was decreased at 2 weeks after the operation compared with the control value, and then gradually increased from 4 to 12 weeks after the operation. The a^* value was increased at 2 weeks but then decreased to a value close to the control value at 12 weeks. The b^* value was not significantly changed at 2 weeks compared with the control value, and then remarkably declined from 4 to 12 weeks (Fig. 1). The spectral curves of all the groups showed two dips at 420 and 560 nm and a specific peak around 490 nm. There was a gradual increase in the spectral reflectance ratio from 620 to 700 nm. Across all the measured wavelengths, there was a low reflectance ratio in group M-2 compared with group C and a gradual increase in the reflectance ratio over the time course after the operation (Fig. 2).

Histomorphological findings

The histological scores described by Caplan et al. [1] were 1.4 in group M-2, 4.9 in group M-4 and 7.3 in group M-12. Significant differences in the scores were seen between groups M-2 and M-4 ($P = 0.007$), between groups M-2 and M-12 ($P = 0.004$) and between groups M-4 and M-12 ($P = 0.01$).

Biochemical measurements

The mean water contents were 84.6% in group M-2, 75.8% in group M-4 and 57.5% in group M-12. Significant differences in the water contents were found between groups M-2 and M-12 ($P = 0.007$) and between groups M-4 and M-12 ($P = 0.004$). The mean hydroxyproline contents were 19.6 nmol/mg in group M-2, 29.3 nmol/mg in group M-4 and 27.6 nmol/mg in group M-12 (Fig. 9B). There were no significant differences among the three groups. The mean chondroitin sulfate contents were 16.7 nmol/mg in group M-2, 44.2 nmol/mg in group M-4 and 18.1 nmol/mg in group M-12. Significant differences in the chondroitin sulfate contents were found between groups M-2 and M-4 ($P = 0.02$) and between groups

M-4 and M-12 ($P = 0.04$). Among the biochemical findings, the water content showed similar change to the results of the a^* value.

Discussion: In the present study, repaired cartilage after a microfracture was evaluated quantitatively using the $L^* a^* b^*$ colorimetric system and the spectral reflectance distribution. Our findings demonstrate the ability of spectrophotometric measurement to judge the repair cartilage after treatment on the basis of objective data such as the L^* , a^* and b^* values and the spectral reflectance curve. Analysis of the many factors affecting cartilage color is a very complex subject that needs much further investigation. Therefore, further studies on the construct validity of the $L^* a^* b^*$ colorimetric system and the spectral reflectance distribution for human articular cartilage are recommended.

References: [1]Caplan AI, et al. Clin Orthop 1997; 342: 254-69.

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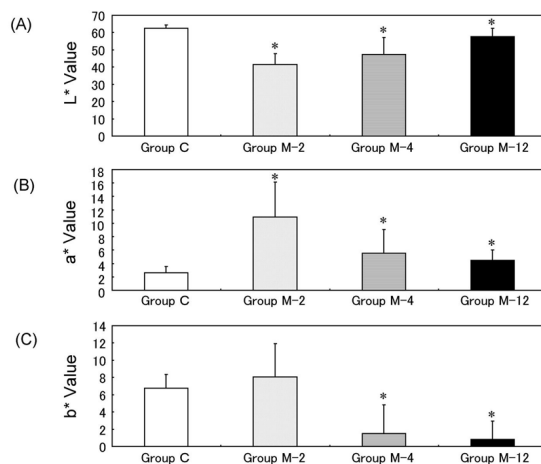


Fig. 1. The L^* , a^* and b^* values of the four experimental models. * $P < 0.05$ vs. the control group (Group C).

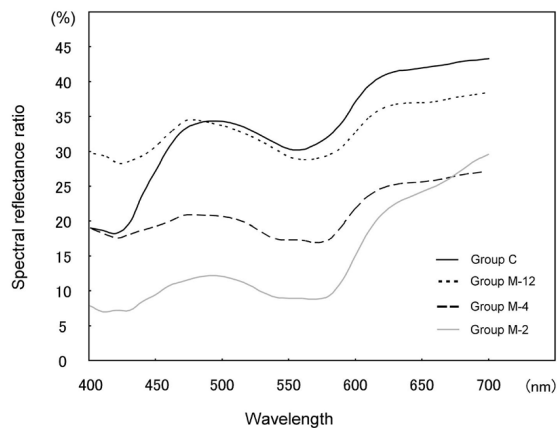


Fig. 2. The spectral reflectance curves of the four groups.