# Rhythmic Changes of Morphometric Parameters of Rat Cardiac Scar Connective Tissue

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**Key Words:** *heart; electrocoagulation; scar connective tissue; morphometric parameters*

Considerable attention has recently been focused on investigations which have found that connective tissue plays an active role in heart and muscle contraction [3-5]. In our previous article [1] we noted that the heart connective tissue, including scar tissue, unlike other organs, has morphological structures with a fibrillar apparatus.

The aim of this study was to attempt to establish the features of the direct involvement of connective tissue scar structures (SS) in cardiac contractility.

## MATERIALS AND METHODS

The experiments were carried out on 12 3-months-old female albino rats weighing 310-312 g. Under ether anesthesia the thorax was opened, and the pericardium was removed. The electrocoagulation of the site in the middle third of the anterior wall of the left ventricle was performed with a Dymok electrocoagulator. All the animals were sacrified 60 days after the operation under ether with thorax opening. The hearts of 4 rats were separated from the major vessels and diastolic cardiac arrest was achieved. Four other hearts were arrested in systole. This was achieved in two ways: 1) two hearts were injected in the sinus venosus with 10% $CaCl_2$ (0.5 mg/kg) until systolic arrest was attained; 2)the other two hearts were cut away from the major vessels and the contracting organ was placed in a 10% $CaCl_2$ solution. In this case cardiac arrest also occurred in systole. The last 4 hearts were cut in two through the connective tissue scar. One part was arrested in diastole and the other in systole, as described above.

Immediately after cardiac arrest, 2-3 1×1 mm pieces were dissected from the scar region and also from the intact region of the left ventricle. The samples were immersed in glutaraldehyde and postfixed in osmium tetroxide. After dehydration, the samples were embedded in Epon. The ultrathin sections, produced with an LKB-8800 ultramicrotome, were double-stained with uranyl acetate and lead citrate. The grids were examined under a UEMV-100V electron microscope.

**TABLE 1.** Morphometric and Stereometric Parameters of Ventricle Cardiomyocytes and Collagen Fibrils of SS of Rat Heart Arrested in Diastole

| Parameter | Results | *p* |
|---|---|---|
| Heart weight | 1137±10.2 | |
| Cardiomyocyte diameter | 14.6±0.25 | <0.001 |
| Relative area of myofibrils | 1.375±0.054 | |
| Myofibril diameter | 0.673±0.077 | <0.001 |
| Collagen fibril diameter | 30.4±2.9 | <0.001 |

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**TABLE 2.** Morphometric and Stereometric Parameters of Ventricle Cardiomyocytes and Collagen Fibrils of SS of Rat Heart Arrested in Systole

| Parameter | Results | *p* |
|---|---|---|
| Heart weight | 1135±9.1 | |
| Cardiomyocyte diameter | 21.4±0.47 | <0.001 |
| Relative area of myofibrils | 1.205±0.123 | |
| Myofibril diameter | 0.986±0.158 | <0.001 |
| Collagen fibril diameter | 28.3±2.5 | <0.01 |

**TABLE 3.** Morphometric and Stereometric Parameters of Ventricle Cardiomyocytes and Collagen Fibrils of SS of Rat Heart, One Part of Which Was Arrested in Diastole and the Other in Systole

| Parameter | Results | | *p* |
|---|---|---|---|
| | Systole | Diastole | |
| Heart weight | 1135±10.4 | – | |
| Cardiomyocyte diameter | 20.1±0.54 | 15.7±0.32 | <0.001 |
| Relative area of myofibrils | 1.483±0.095 | 1.506±0.049 | |
| Myofibril diameter | 0.993±0.147 | 0.577±0.084 | <0.001 |
| Collagen fibril diameter | 27.8±1.8 | 33.1±2.7 | <0.01 |

The remaining heart was fixed in 10% formalin and embedded in paraffin. Sections 5-7μ thick were stained with hematoxylin and eosin and after van Gieson. The following morphometric parameters were measured: weight of body (g); absolute weight of heart (mg); cardiomyocyte diameter (μ); relative area of myofibril surface ($m^2/cm^3$).To assess the morphometric parameters of the scar collagen fibrils, the diameters of 100 collagen fibrils (nm) were measured for every case, and the arithmetic mean was calculated. We were guided by the recommendations presented in monograph [2] when choosing the morphometric and stereometric measurements.

Statistical analysis of the findings was performed according to the laws of variational statistics.

## RESULTS

The histological features of the SS were identical to those described previously [1]. The SS was composed of fibrillar structures with relatively few blood vessels and cells (mainly elongated fibroblasts and fibrocytes). Myogenic elements were obtained in all the experimental series [1]. The morphometric and stereometric data of the myocardial structures and SS are presented in Tables 1-3.

As can be seen from the above results, during heart contraction, along with cardiomyocyte contraction there is a simultaneous extension of the collagen fibers of the SS by approximately 10-15%. In diastole the collagen fibers return to their original state. The findings evidently call for further investigation and serious analysis because there are a number of unresolved questions for which there are no immediate answers. However, based on the experimental results, it may be concluded that the SS is not functionally dead and takes part in the contractile activity of the heart as an integral organ.

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