FUNCTIONAL AND STRUCTURAL DIFFERENCES IN FIBROBLASTS FROM ATRIA OF PATIENTS WITH AND WITHOUT ATRIAL FIBRILLATION

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BACKGROUND
Cardiac chamber-specific differences in the responsiveness of fibroblasts to pathogenic stimuli have been described in animal models. However, it is unclear whether disease states affect the structure and function of cardiac fibroblasts, especially in the human heart.

OBJECTIVE
Our objective of this study was to determine functional and structural differences in fibroblasts isolated from atrial appendages of patients with and without atrial fibrillation.

METHODS
Fibroblasts from human atrial appendage tissue removed during open heart surgery from patients with and without atrial fibrillation (AF) were grown in culture. After 14 days, the outgrown cells were detached, counted, and then grown in culture to passage 3 and used at this passage. Imaging of the attached cells, scratch assay, and western blot EdU incorporation assay was performed to determine the differences in fibroblast size, structure, motility as well as time, and efficiency of wound closure and activation to myofibroblasts. Total cell protein was also determined at the same time.

The differences in fibroblast size, structure, motility as well as time, and efficiency of wound closure and activation to myofibroblasts were compared between fibroblasts from AF and non-AF patients using student t test and p<0.05 was considered as significantly different.

RESULTS

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<th>Clinical Characteristics of Patients</th>
<th>Morphology of Primary Cultured Fibroblasts</th>
<th>EdU Incorporation and Proliferation Difference of Fibroblasts</th>
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Overall Yield of Fibroblasts and Protein from atrial tissue from Non-AF and AF patients

**Figure 1.** A. Number of fibroblasts obtained at day 14 from the atrial tissue of patients with non-AF (n=8) and AF (n=8). B. Total protein obtained from 10^6 fibroblasts from Non-AF and AF patients (n=8) at passage 3 culture. (* p<0.05)

**Figure 2.** Calcein-AM treated cells outlining the cell morphology of fibroblasts from Non-AF and AF patients.

**Figure 3.** A. Scratch Assay over a span of 6 hours between Non-AF and AF fibroblasts. B. Data points show fibroblasts from Non-AF and AF patients with a unidirectional slope (n=8). (* p<0.01)

**Figure 4.** A. Bar graph of motility differences between control fibroblasts and fibroblasts treated with CTD from Non-AF and AF patients. B. EdU incorporation determined by western blot from the Non-AF and AF fibroblasts.

**Figure 5.** A. EdU incorporation assay determining proliferation differences between Non-AF and AF fibroblasts over a span of 24 hours. B. Quantitative data of EdU incorporation shown in bar graph form.

CONCLUSIONS
This study, for the first time, identifies differences in fibroblasts from human atria between AF and non-AF patients with regard to size, shape, motility, time to close wound and proliferation rate.

Further investigation of functional significance of these differences in fibroblasts on cardiac repair after injury and progression of AF and its complications are warranted.

DISCLOSURE
All authors have no financial conflict of interest related to this study.