# Stretch, stiffness, sensing, signaling & speed

# Impact

The main goal of this thesis was to investigate mechanisms of mechanosensing of cardiac fibroblasts (CFs). To this end we examined the effect of mechanical stimulation in the form of cyclic stretch on CFs, using *in vitro* models and, to study the effect of long-lasting stretch, in an *in vivo* setting of mitral regurgitation in dogs.

**Scientific impact**

CFs are important during healing after myocardial infarction and in the adaptation of the heart to increased mechanical load. While the repair function of fibrosis by CFs after myocardial injury is clearly beneficial, excess fibrosis causes a stiffer myocardium and conduction disorders, contributing to development of cardiac dysfunction and arrhythmia. Better understanding of the fibrotic processes may help finding ways to avoid the negative and take benefit of positive parts of the fibrotic processes.

Fibrosis, characterized by excess matrix protein deposition, occurs when CFs differentiate into myofibroblasts under the influence of different stimuli like TGFβ1 and mechanical loading.

In this thesis we have unveiled a mechanism of mechanosensing and regulation of CFs involving the mechanosensitive ion channel Piezo1 in both stretch-induced upregulation of the brain natriuretic peptide (BNP) (generally known for expression by cardiomyocytes and as plasma marker of heart failure) and the stretch-induced downregulation of Cilp1 (chapters 2 and 3). These findings may open new doors for pharmacological modulation of fibrosis, for example by stimulation of Piezo1.

Such future studies may also benefit from another part of our research: the development of a novel 3D culture system of Engineered Heart Matrix (EHM), composed of CFs cultured within a collagen-1 matrix gel. We showed that it is possible to culture CFs within the 3D EHM with a stiffness similar to that of normal healthy myocardium and that the use of such physiological stiffness results in a more quiescent CF state when compared with CFs cultured on commonly used hard plastic culture plates.

**Societal impact**

Heart failure forms a large social and economic burden, because it, accounts for 19% of all cardiac deaths and it required frequent hospitalizations, creating costs of half a billion euros per year in the Netherlands. Fibrosis, involving modifications of CFs, is an important factor in the development of heart failure.

Like in many other research groups, most of the CFs used in cell cultures were derived from rats, therefore involving animal experiments. During the last part of the PhD period we used Induced Pluripotent Stem Cells (iPSC's). These are human derived cells and therefore

creates opportunity to replace animal experiments. Moreover, the human nature of these cells likely increases the clinical relevance of the studies. Moreover, when the cells are taken from patients it also opens the door towards investigating patient specific treatment. The aforementioned Engineered Heart Matrix seems ideal to perform such studies with human iPSC's.