Pleiotropic roles of the matricellular protein Sparc in tendon maturation and ageing.

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FIGURE LEGENDS: Supplementary figures

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Figure S1. Differential gene expression in mouse Achilles tendon tissue

11 (a) Quantitative RT-PCR results demonstrating repression of Matrix metalloproteinases 2 and 9 and

the tendon cell associated marker Mohawk in healthy-aged mouse Achilles tendons. A change in

expression ≥2.0 was assumed as a significant change (n=5 Achilles tendon samples); graphs represent

means±SEM. (b) Repression of the above-mentioned genes is also seen in in tendon progenitor/stem

cells of healthy-aged mouse Achilles tendons in vitro (passage nr. 3). qPCR data for healthy-aged

tendons were normalized to young tendons, a fold change ≥ 2.0 was assumed as a significant difference

in expression; shown are means±SEM.

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Figure S2. Collagen type I to type II mRNA ratio

- Comparison of collagen type I to type III mRNA ratios for young, Sparc-/-, and healthy-aged Achilles
- 21 tendons. Data represent means±SEM of 3 independent biological replicates; ***P < 0.001, One-way
- 22 ANOVA (Tukey's Multiple Comparison Test).

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Figure S5. Cell area and cell migration

- 25 (a) Cell area of Sparc-/- TDSPCs on fibronectin coated surfaces is significantly reduced compared to
- young and healthy-aged wild type cells. No significant differences between young and healthy-aged
- 27 tendon cells are seen. Cell area of a minimum of 160 cells was measured, ***P < 0.001, **P < 0.01, n.s.
- = not significant, (Kruskall-Wallis test with Dunn's Multiple Comparison test). (b, c) Wound healing
- assays on collagen type I and fibronectin coated surfaces reveal moderate changes in cell migration

- 30 speed in vitro for Sparc-/- and healthy-aged cells migrate moderately faster in vitro. Dot blots show
- 31 results from 3 independent experiments and red bar indicates mean. Representative images for scratch-
- wound assays 0h and 10h after creating the wound are shown. ***P < 0.001, **P < 0.01, One-way
- 33 ANOVA (Tukey's Multiple Comparison Test).

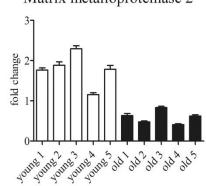
2 **Supplementary Figures**

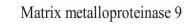
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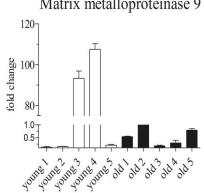
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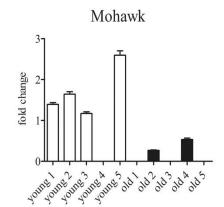
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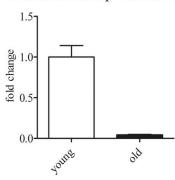




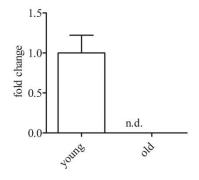




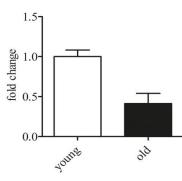
Matrix metalloproteinase 2



Matrix metalloproteinase 9



Mohawk

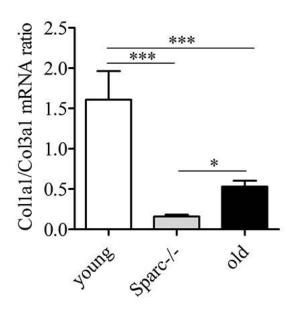


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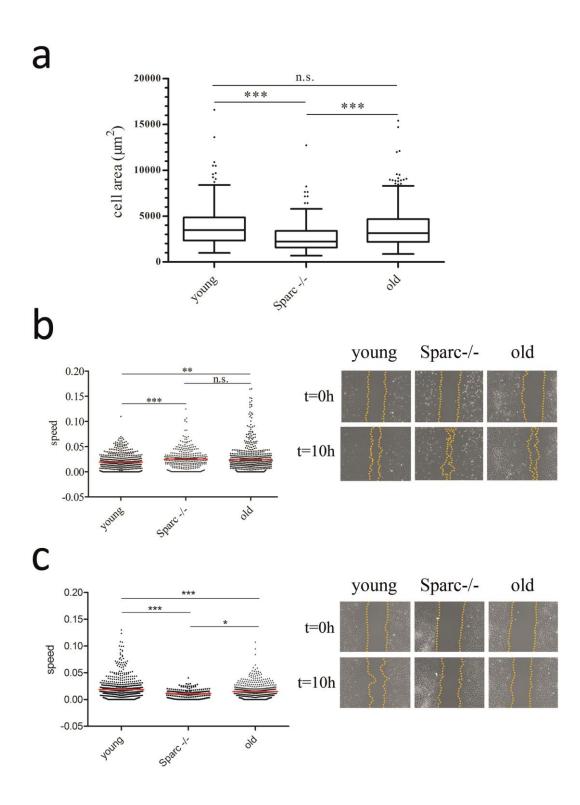
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Suppl Figure 1



9 Suppl Figure 2



Suppl Figure 5