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Supplemental Information

Topical Lyophilized Targeted Lipid Nanoparticles in the Restoration of Skin Barrier Function following Burn Wound

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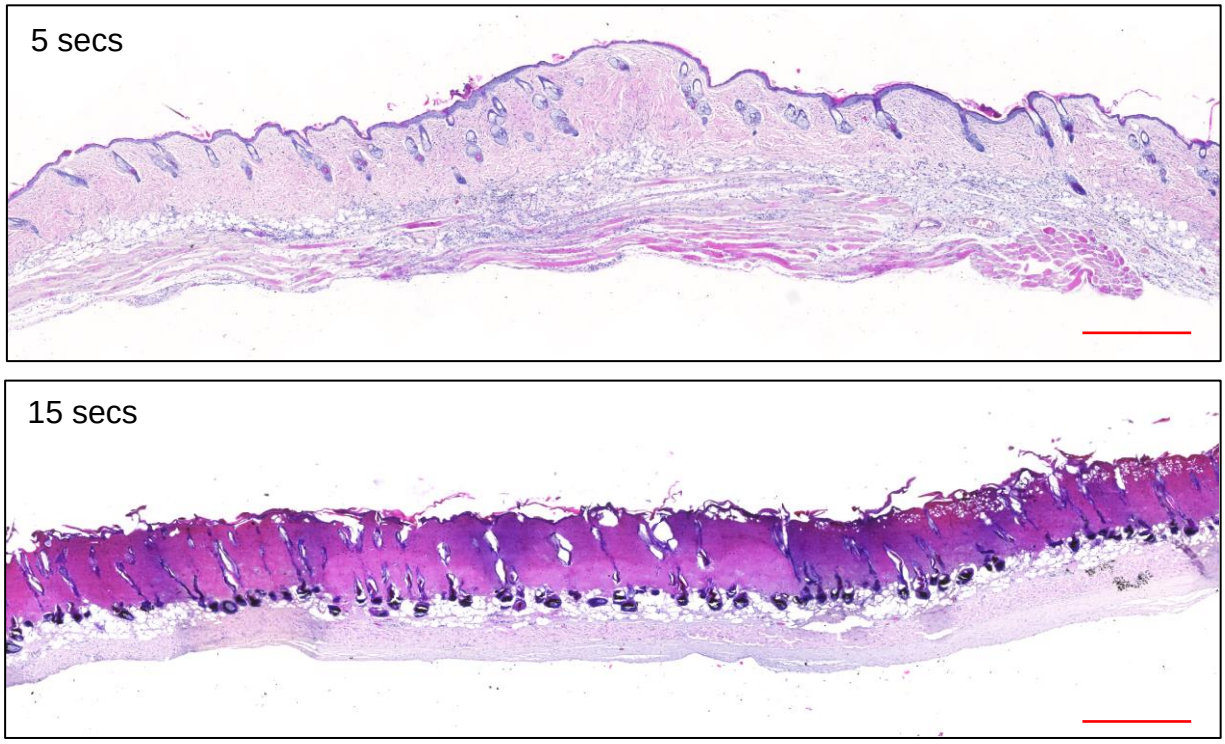
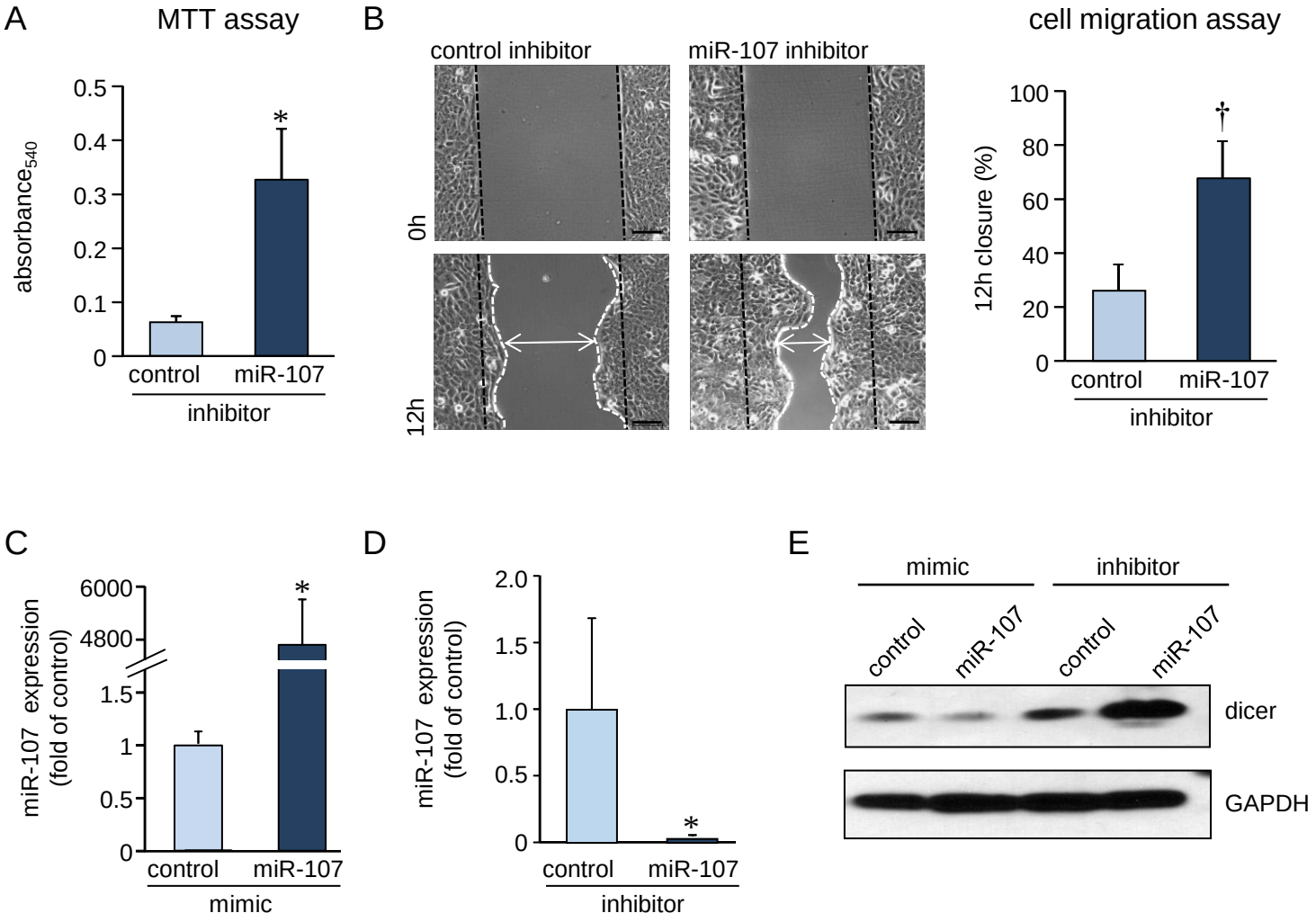
A



Supplementary Figure S1: (A) Representative mosaic image of murine dorsal skin after application of the burner for 5 secs (top) and 15 secs (bottom) for developing the full thickness burn wound. Scale bar = 500µm.

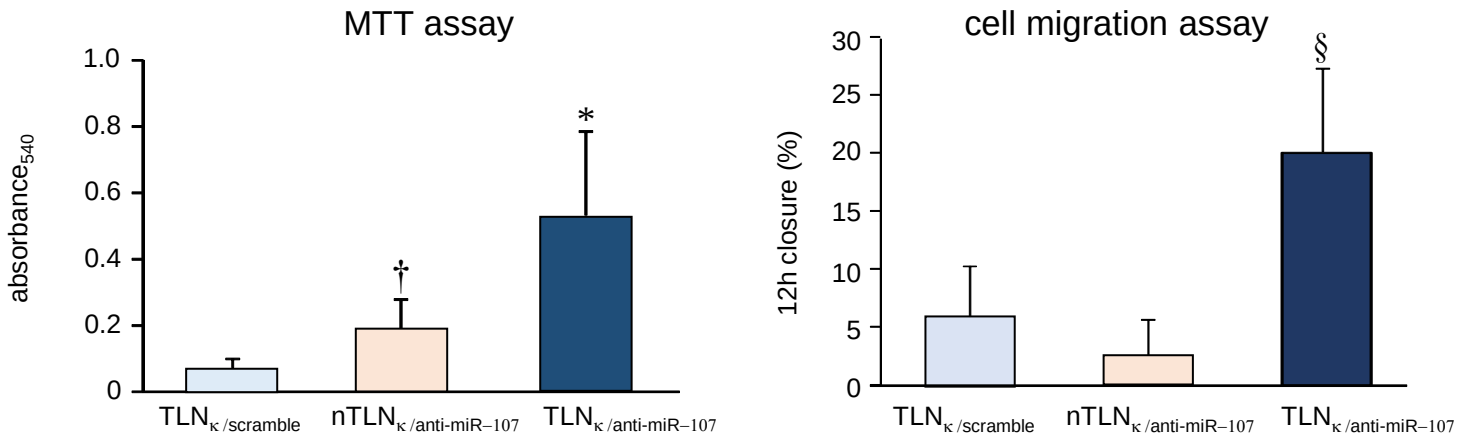
Figure S1



Supplementary Figure S2: HaCaT cells were transfected with either control inhibitor or miR-107 inhibitor for 72h. **(A)** Cells were trypsinized and reseeded in 96 wells plate. Cell proliferation assay was performed after 24h using MTT assay. Data expressed as mean ± SD. * $p<0.001$; n=8 **(B)** Cell migration assay was performed after reseeding in 2-well cell inserts. Migration of cells was observed at 12h following removal of the insert. The black and white dashed line indicated the distance at 0h and 12h respectively. Scale bar = 100 mm. The distance between the two ends are calculated using Zen software (Zeiss) and expressed graphically. Data expressed as mean ± SD. † $p<0.01$; n=3. Quantitative PCR analysis of miR-107 after delivery of **(C)** miR-107 mimic and **(D)** miR-107 inhibitor in HaCaT cells. Data expressed as mean ± SD (n=4). * $p<0.001$. **(E)** Western blot analysis of dicer expression after transfection of miR-107 mimic and inhibitor.

Figure S2

A


MTT assay
absorbance540
1.0
0.8
0.6
0.4
0.2
0
†
*
TLNκ /scramble
nTLNκ /anti-miR−107
TLNκ /anti-miR−107
cell migration assay
12h closure (%)
30
25
20
15
10
5
0
§
TLNκ /scramble
nTLNκ /anti-miR−107
TLNκ /anti-miR−107

B

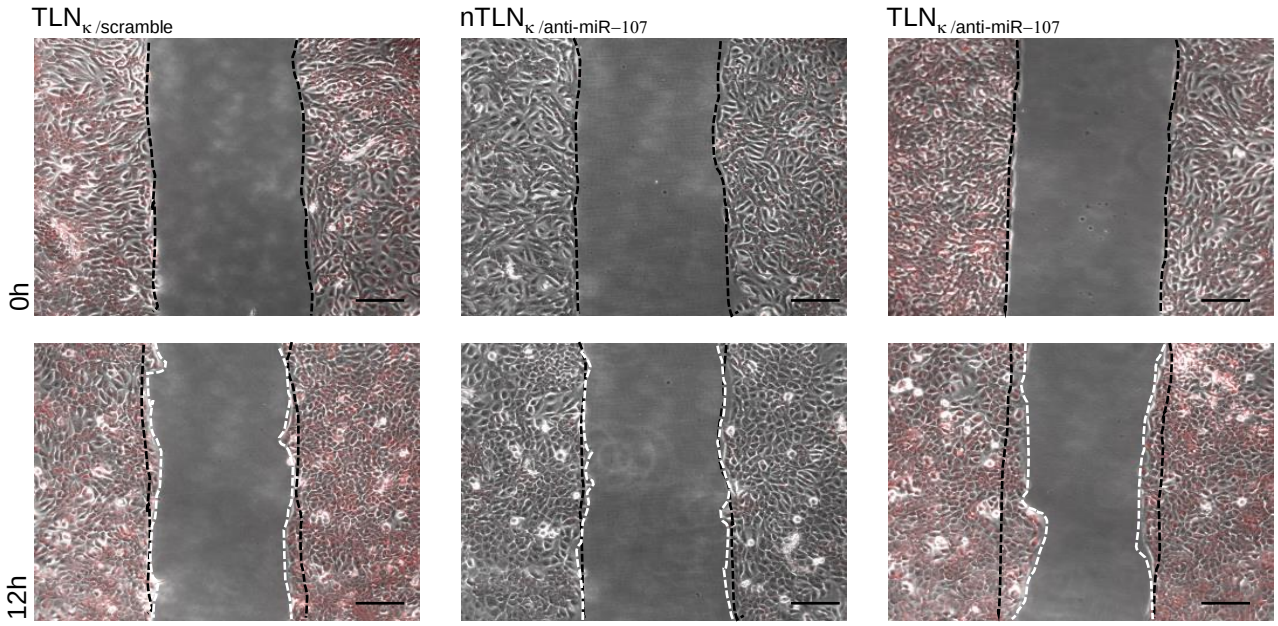
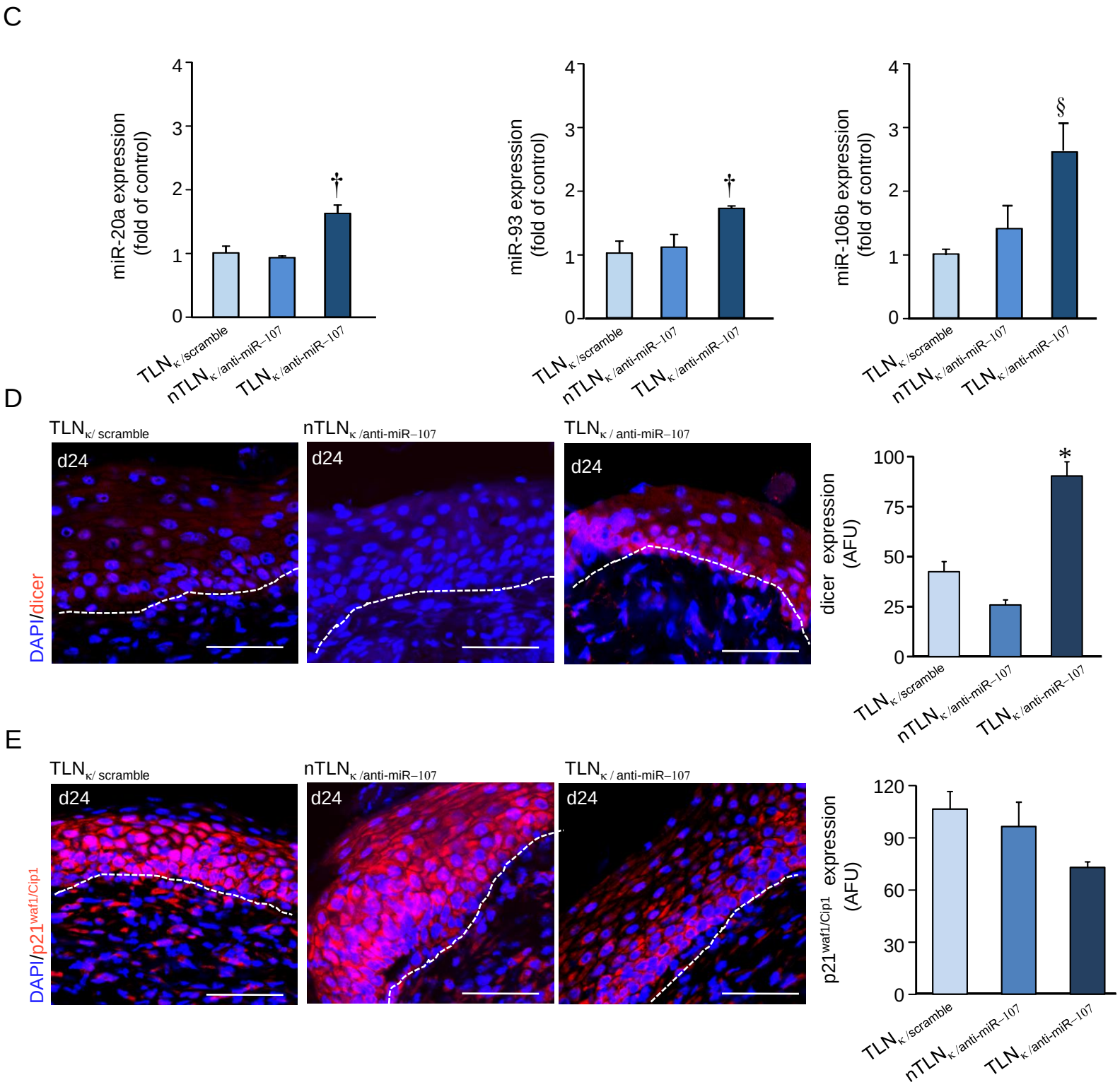
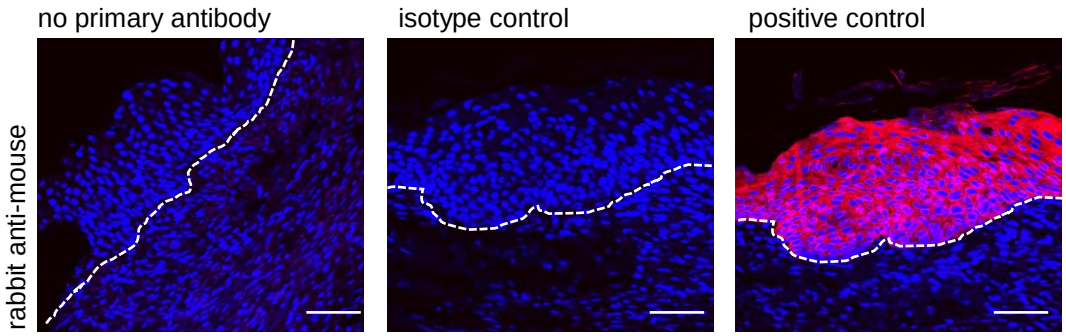

TLNκ /scramble
nTLNκ /anti-miR−107
TLNκ /anti-miR−107
0h
12h

Figure S3



Supplementary Figure S3: HaCaT cells were seeded in either 96 well plate of 2-well inserts and treated with either $TLN_{\kappa/scramble}$, $nTLN_{\kappa/anti\text{-}miR\text{-}107}$ and $TLN_{\kappa/anti\text{-}miR\text{-}107}$. The LNPs were tagged with DiD (red). **(A)** Cell viability was determined after 24h of treatment by using MTT assay . Data expressed as mean ± SD. * p<0.001; n=8 **(B)** Migration of cells was observed at 12h following removal of the insert. The black and white dashed line indicated the distance at 0h and 12h respectively. Scale bar=100 mm. The distance between the two ends are calculated using Zen software (Zeiss) and expressed graphically. Data expressed as mean ± SD. § p=0.0544; n=3. **(C)** Quantitative PCR analysis of miR-20a, miR-93 and miR-106b at day 24 after delivery of $TLN_{\kappa/scramble}$, $nTLN_{\kappa/anti\text{-}miR\text{-}107}$ and $TLN_{\kappa/anti\text{-}miR\text{-}107}$. Data expressed as mean ± SEM (n=3). § p<0.05; † p<0.01.$TLN_{\kappa/anti\text{-}miR\text{-}107}$ increased the expression of **(D)** dicer (red), and (**E**) decreased the expression of $p21^{waf1/Cip1}$ in murine wound-edge at day 24. Sections were counter stained with DAPI. Dermal-epidermal junction is indicated by dashed white line. Scale bar=50µm. Abundance of dicer and $p21^{waf1/Cip1}$ in epidermis were quantified and expressed graphically as mean ± SEM. (n=3).* p<0.001, ANOVA.

Figure S3



Supplementary Figure S4: Specificity of the antibodies used in the study was validated using no antibody control and isotype controls of host species. The white dash line indicates the epidermal and dermal junctions. Scale bar = 50μm.

Figure S4