

SUPPLEMENTARY MATERIAL

SUPPLEMENTARY FIGURES

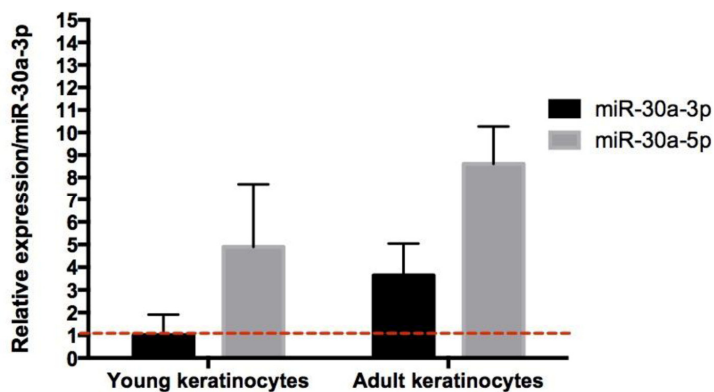


Figure S1. Relative expression of miR-30a-3p and miR-30a-5p in human keratinocytes. MiR-30a-3p and miR-30a-5p expression levels were measured by q-PCR on cultured keratinocytes from young and aged donors. Data were calculated using miR-30a-3p expression levels as a reference. Results are mean +/- SD from three independent samples.

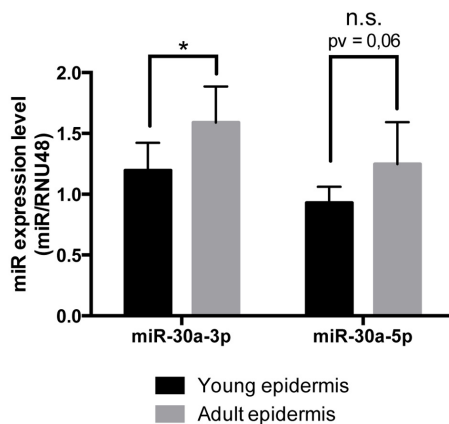


Figure S2. Relative expression of miR-30a-3p and miR-30a-5p in in young and aged epidermis. MiR-30a-3p and miR-30a-5p expression levels were measured by q-PCR on epidermis RNA samples from young and aged donors. Data were calculated using miR-30a-5p expression levels as a reference. Results are mean +/- SD from three independent samples. *P<0,05.

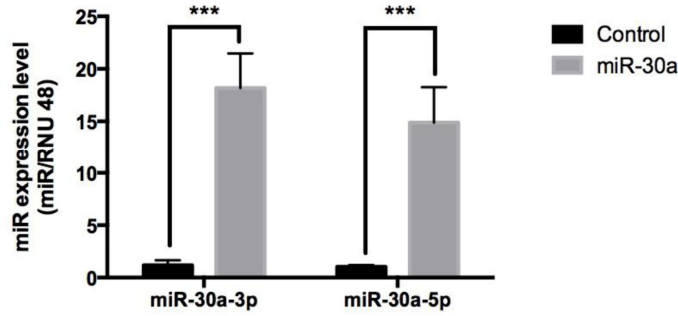


Figure S3. MiR-30a-3p and miR-30a-5p level in REs overexpressing miR-30a. Keratinocytes were infected with lentiviral vector particules of pSLIK-Venus control or pSLIK-Venus miR-30a, treated by doxycyclin and then used to generate reconstructed epidermis. MiR-30a-3p and miR-30a-5p expression levels were measured on total REs RNA sample by quantitative-PCR. Results are mean +/- SD from three independent samples. ***P<0,001.

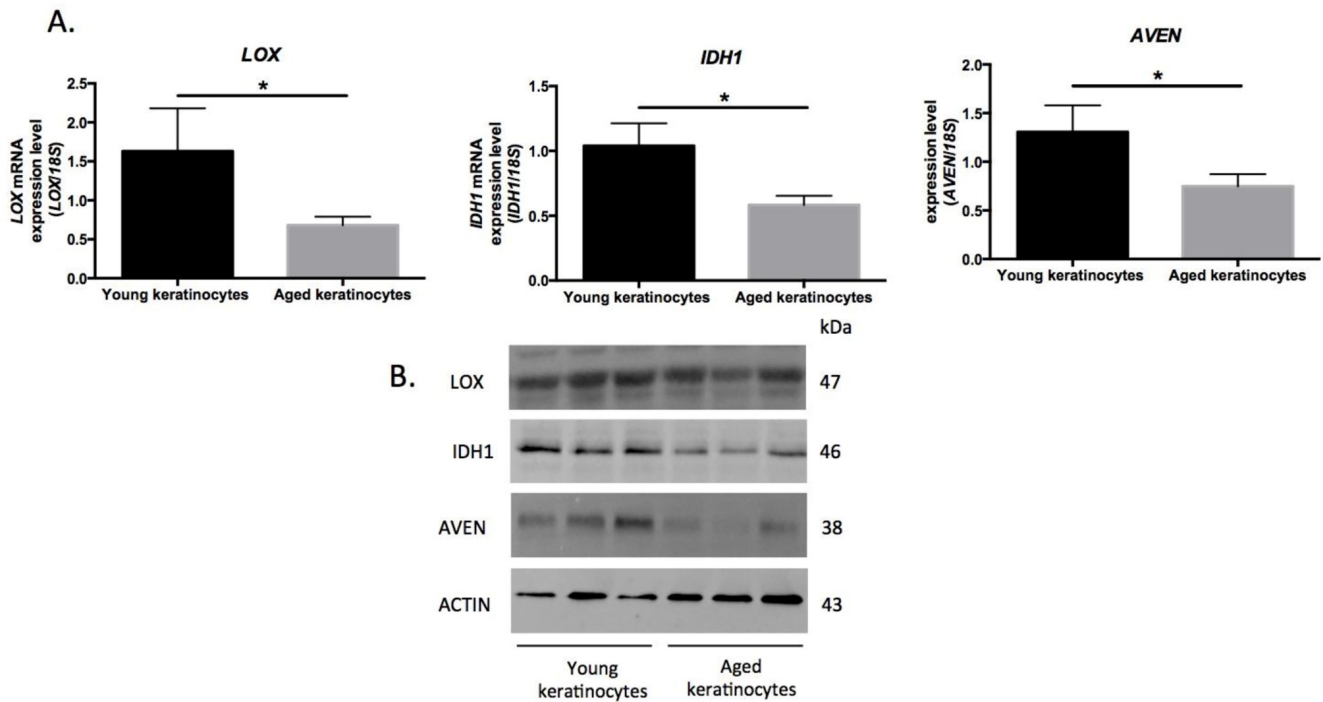


Figure S4. LOX, IDH1 and AVEN expression in keratinocytes from young and aged skin. (A) The expression level of AVEN, IDH1 and LOX transcripts were evaluated by qPCR in RNA samples from keratinocytes prepared from young or aged skin. Results are mean +/- SD from 8 independent samples. *P< 0,05. **(B)** The expression levels of AVEN, IDH1 and LOX proteins were evaluated by western blotting in cultured keratinocytes prepared from young or aged skin. Actin was used as a loading control.

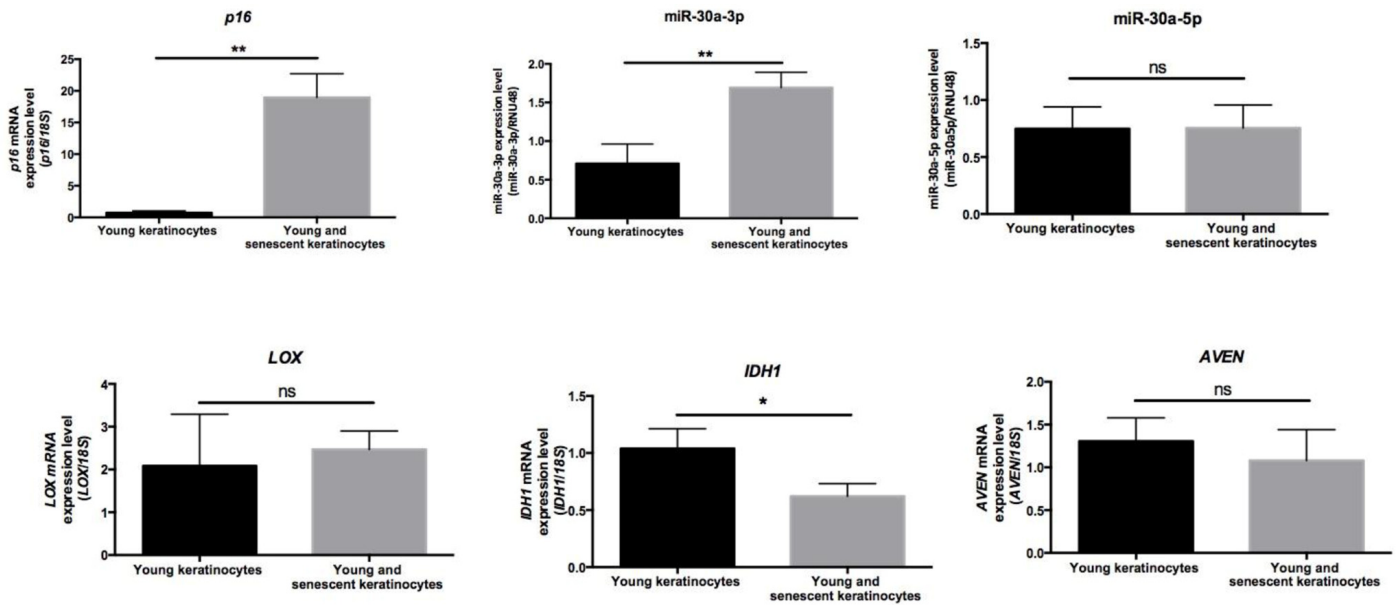


Figure S5. P16, miR-30a-3p, miR-30a-5p, LOX, IDH1 and AVEN expression in normal and replicative senescent keratinocytes from young skin The expression level of miR-30a-3p, miR-30a-5p, AVEN, IDH1 and LOX transcripts were evaluated by qPCR in RNA samples from keratinocytes prepared from young skin at early passage (passage 2) and replicative senescent keratinocytes (passage 6). P16 was used as a marker of senescence. Results are mean +/- SD from three independent samples. *P< 0,05. **P< 0,01. ns : non significant.

SUPPLEMENTARY MATERIALS AND METHODS

List of primers used for QPCR

Gene	Gene ID	Primer	Primer Sequence
AVEN	57099	Reverse 5'	GCCAAGCTTGCATGCCTTGTTCATCTA-3'
		Forward 5'	ATGAGCTCGCAGCTTGTTCATTTGTTT-3'
IDH1	3417	Reverse 5'	GCCAAGCTTCATGTTACAAAGGTGGCAAT -3'
		Forward 5'	ATGAGCTCTCAAAGCTCAGGCCAAA-3'
LOX	4015	Reverse 5'	TTGGTCGGCTGGGTAAGAAAT-3'
		Forward 5'	GGATACGGCACTGGCTACTTC-3'
18S	100008588	Reverse 5'	CGATGCGGCGGCGTTATT-3'
		Forward 5'	CCTGGTGGTGCCCTTCCGT-3'

List of primers used for the amplification of the target genes 3'-UTR

Primers	Restriction enzyme	Sequences
3' UTR AVEN forward	SacI	5'- ATgagctcGCAGCTTGTTCATTTGTTT-3'
3' UTR AVEN reverse	HindIII	5'-GCCaagcttGCATGCCTTGTTCATCTA-3'
3' UTR IDH1 forward	SacI	5'-ATgagctcTCAAAGCTCAGGCCAAA-3'
3' UTR IDH1 reverse	HindIII	5'-GCCaagcttCATGTTACAAAGGTGGCAAT-3'
3' UTR LOX forward	SacI	5'-ATgagctcATGGACACATCTGGTGCTGA-3'
3' UTR LOX reverse	HindIII	5'-GCCaagcttCTGCCCATGGGAAAGATAAAA-3'

List of primers used for the mutagenesis of the potential miR-30a MRE

Primers	Sequences
mutAVEN forward	5'-CAACATAGCCAGTGTCAGCATAGCAGATGC-3'
mut AVEN reverse	5'-GCATCTGCTATGCTGACACTGGCTATGTTG-3'
mut IDH1 f - site 1	5' GGTAAGTGGTCTACAGGTCATTTTTCTGTGTTACAC-3'
mut IDH1 r - site 1	5'-GTGTAACACAGAAAAATGACCTGTAGACCTAGTTACC-3'
mut IDH1 f- site 2	5'-GAGTTTATCTTTTCTATAAGTCAGCCTTTTCTTATATATAC-3'
mut IDH1 r - site 2	5'-GTATATATAAGAAAAAGGCTGACTTATAGAAAAGATAAACTC-3'
mut LOX forward	5'-CCCTATATAAAAAGTATGTCATTTAAAAAATTAGTAG-3'
mut LOX reverse	5'-CTACTAATTTTTTAAATGACATACTTTTTATATAGGG-3'