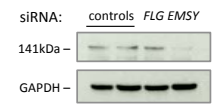
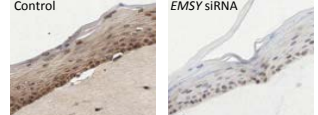
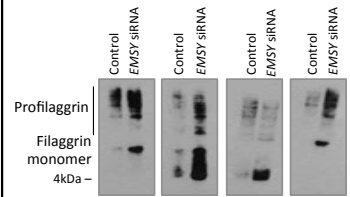
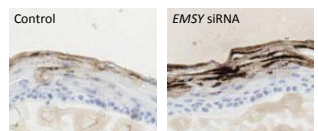
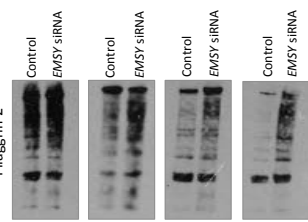
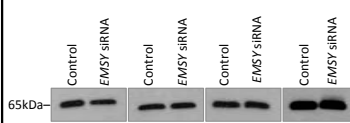
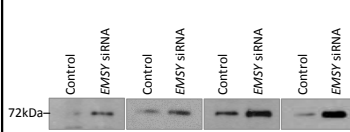


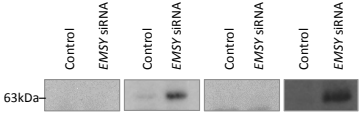
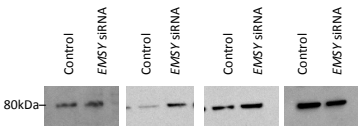
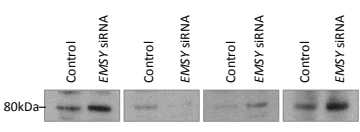
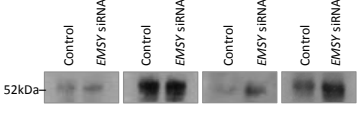
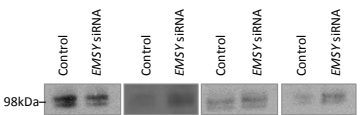
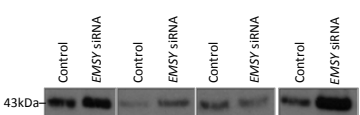
SUPPLEMENTARY FIGURES AND TABLES - continued

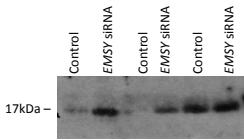
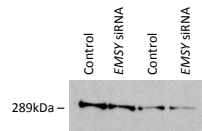
***EMSY* expression affects multiple components of skin barrier with relevance to atopic dermatitis**

Martina S Elias PhD,^{1*} Sheila C Wright HNC,¹ Judit Remenyi PhD,¹ James C Abbott PhD², Susan E Bray PhD³, Christian Cole PhD², Sharon Edwards MBChB⁴, Marek Gierlinski PhD², Mateusz Glok¹, John A McGrath FRCP⁵, William V Nicholson PhD¹, Lavinia Paternoster PhD⁶, Alan R Prescott PhD⁷, Sara Ten Have PhD⁸, Phillip D Whitfield PhD⁹, Angus I Lamond PhD⁸ and Sara J Brown FRCPE^{1,10*}

Fig E8. qPCR, Western blotting and immunofluorescence to test for validation of selected protein expression changes identified by mass spec analysis

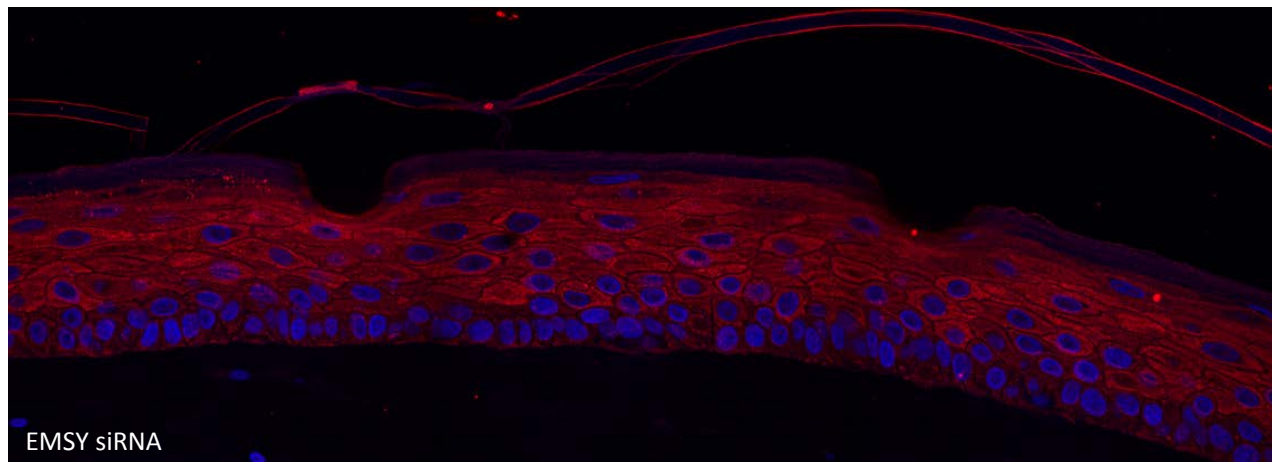
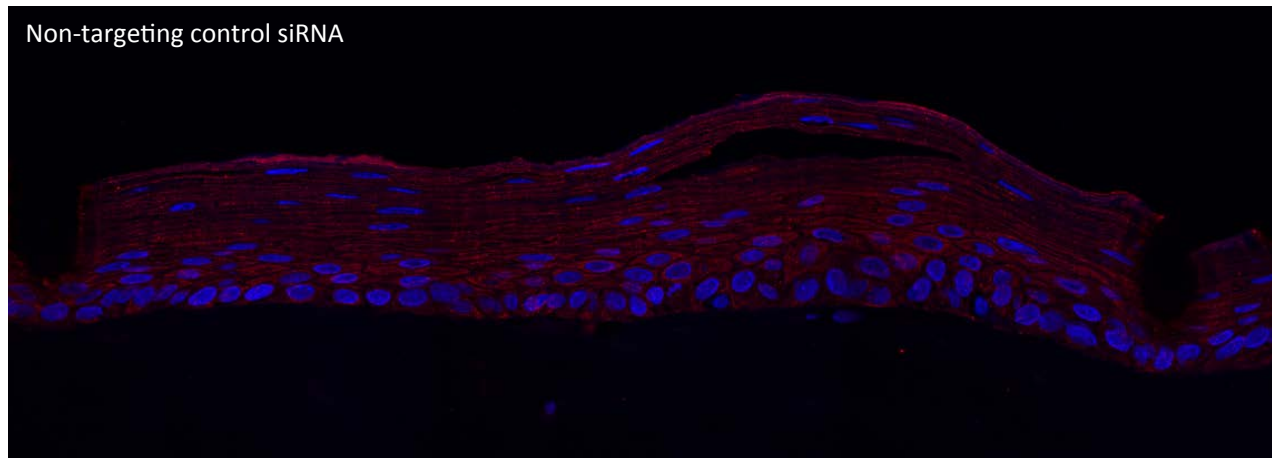
Gene	Protein	Mean FC in m/s proteomics (n=4)	Function	qPCR (fold change n=4, mean \pm SEM)	Western blotting (representative images from ≥ 4 bio-replicates)	Immunostaining (representative images from ≥ 3 bio-replicates)
<i>EMSY</i>	EMSY	Below threshold for detection in mass spec analysis	Transcriptional regulator	0.68 ± 0.04		
<i>FLG</i>	Filaggrin	12.5	Structural and multifunctional	5.07 ± 4.78		
<i>FLG2</i>	Filaggrin 2	38.1		25.28 ± 19.57		Not done
<i>KRT2</i>	Keratin 2	7.9		41.90 ± 34.94		Immunofluorescence images shown following this table
<i>HAL</i>	Histidine ammonia lyase	11.9		17.92 ± 14.23		Immunofluorescence images shown following this table
<i>BLMH</i>	Bleomycin hydrolase	5.8	Filaggrin processing	8.50 ± 2.78	Not done	Not done
<i>ASPRV1</i>	Retroviral-like aspartic protease 1	11.8		43.92 ± 36.25	Not done	Not done

Gene	Protein	Mean FC in m/s proteomics (n=4)	Function	qPCR (fold change n=4, mean ± SEM)	Western blotting (representative images from ≥4 bio-replicates)	Immunofluorescence (representative images from ≥3 bio-replicates)
<i>STS</i>	Steroid sulphatase	7.3	Lipid processing and metabolism	1.25 ± 0.45		Nonspecific staining
<i>ALOXE3</i>	Arachidonate lipoxygenase 3	9.7		4.84 ± 1.25		Nonspecific staining
<i>ALOX12B</i>	Arachidonate lipoxygenase 12	7.4		5.43 ± 2.61		Immunofluorescence images shown following this table
<i>APOE</i>	Apolipoprotein E	3.1		1.87 ± 0.36	Not done	Not done
<i>CDSN</i>	Corneodesmosin	51.6	Cell-cell adhesion	56.38 ± 50.78		Immunofluorescence images shown following this table
<i>DSC1</i>	Desmocollin 1	7.2		2.90 ± 0.47		Immunofluorescence images shown following this table
<i>GJA1</i>	Gap junction alpha-1 protein (connexin 43)	2.5	Cell-cell communication	1.21 ± 0.27		Nonspecific staining

Gene	Protein	Mean FC in m/s proteomics (n=4)	Function	qPCR (fold change n=4, mean ± SEM)	Western blotting (representative images from ≥4 bio-replicates)	Immunofluorescence (representative images from ≥3 bio-replicates)
<i>IL36RN</i>	Interleukin-36 receptor antagonist protein	4.2	Control of inflammation	6.70 ± 4.03		Nonspecific staining
<i>mTOR</i>	Mechanistic target of rapamycin	0.3	Cell cycle and cell proliferation	1.36 ± 0.25		Immunofluorescence images shown following this table
<i>CDK1</i>	Cyclin-dependent kinase 1	6.8	Cell cycle	1.32 ± 0.21	Not done	Not done
<i>COL7A1</i>	Collagen alpha-1 7 chain	5.3	Basement membrane	1.46 ± 0.53	Not done	No clear staining seen
<i>RNase7</i>	Ribonuclease 7	20.5	Antimicrobial	17.76 ± 15.13	Not done	Not done

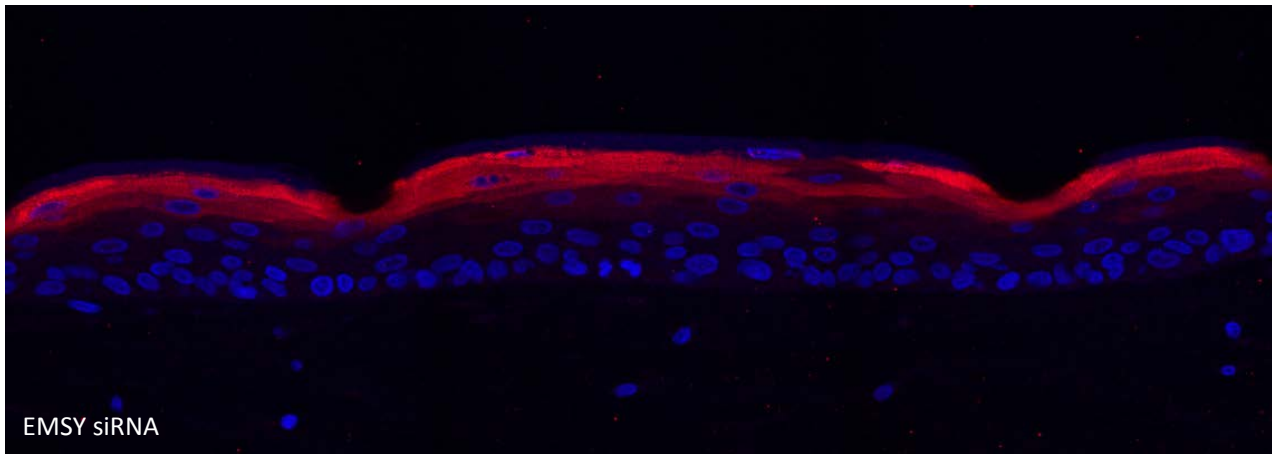
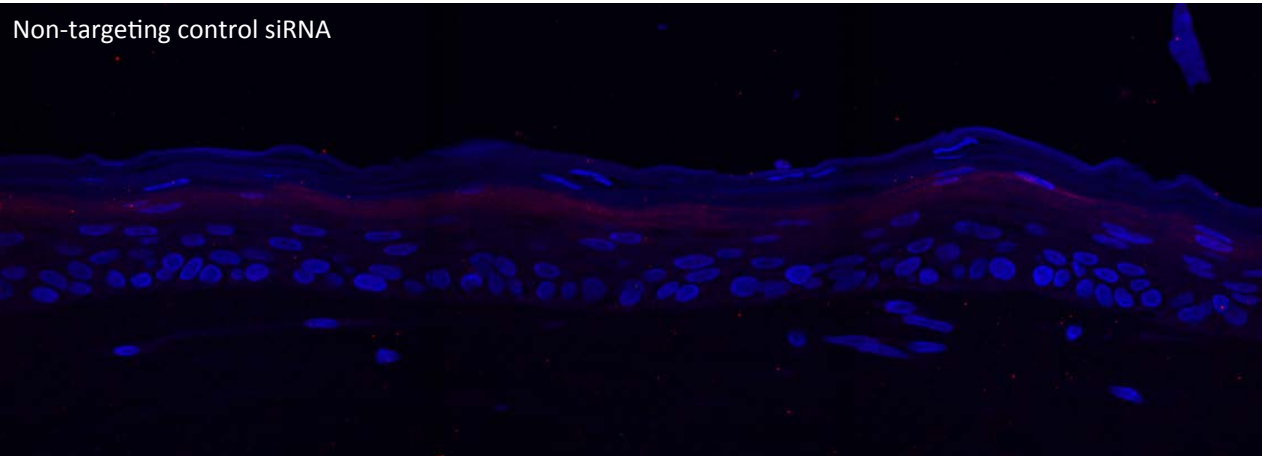
Keratin 2 DAPI

Composite image created by Zeiss LSM710 microscope



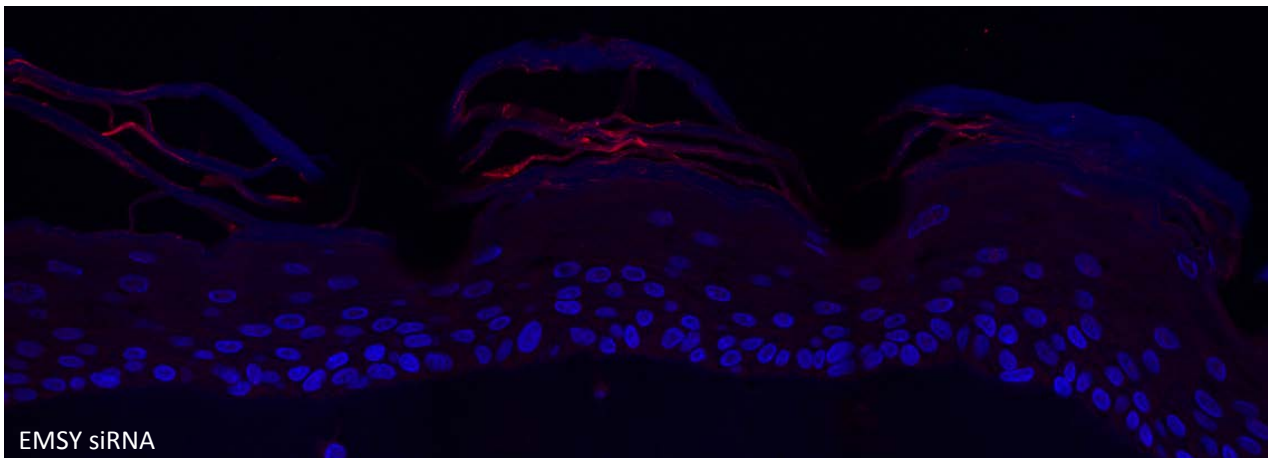
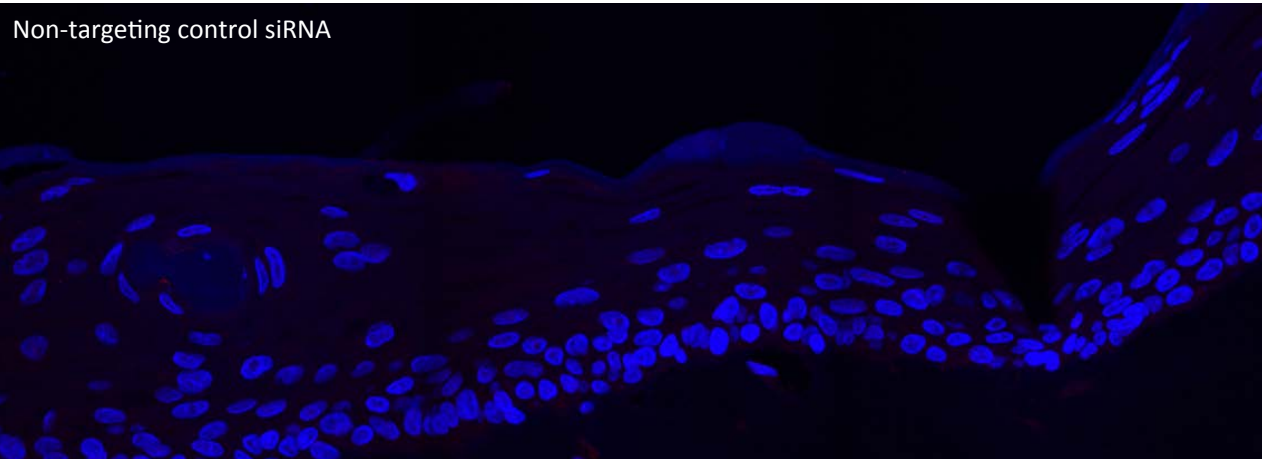
Histidine ammonia lyase DAPI

Composite image created by Zeiss LSM710 microscope

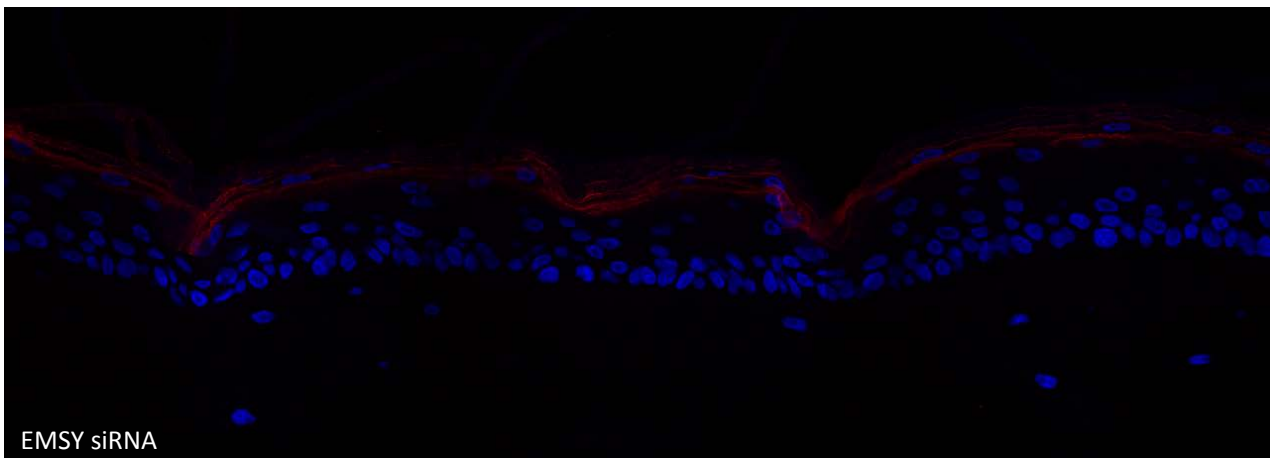
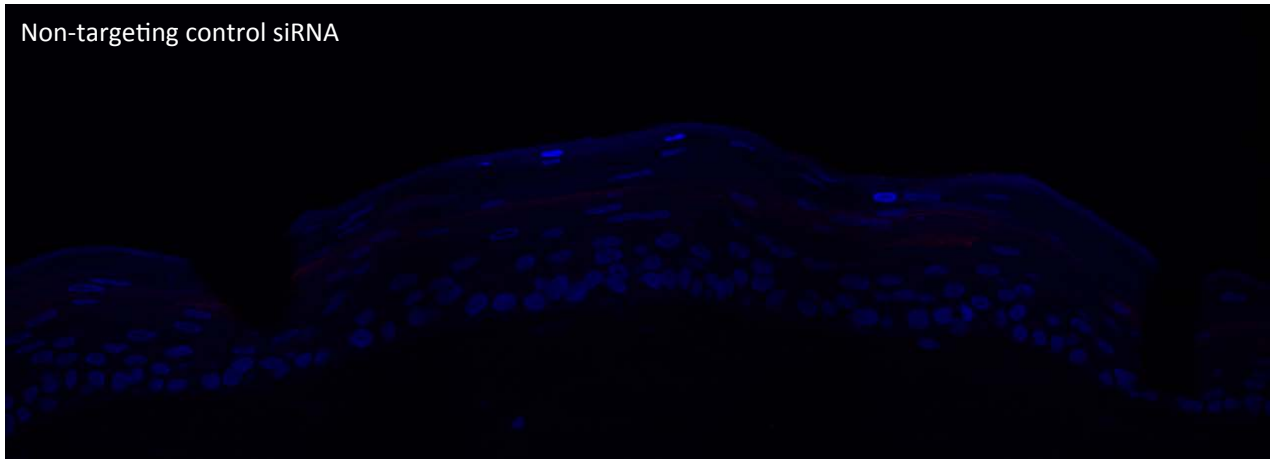


ALOX12 DAPI

Composite image created by Zeiss LSM710 microscope

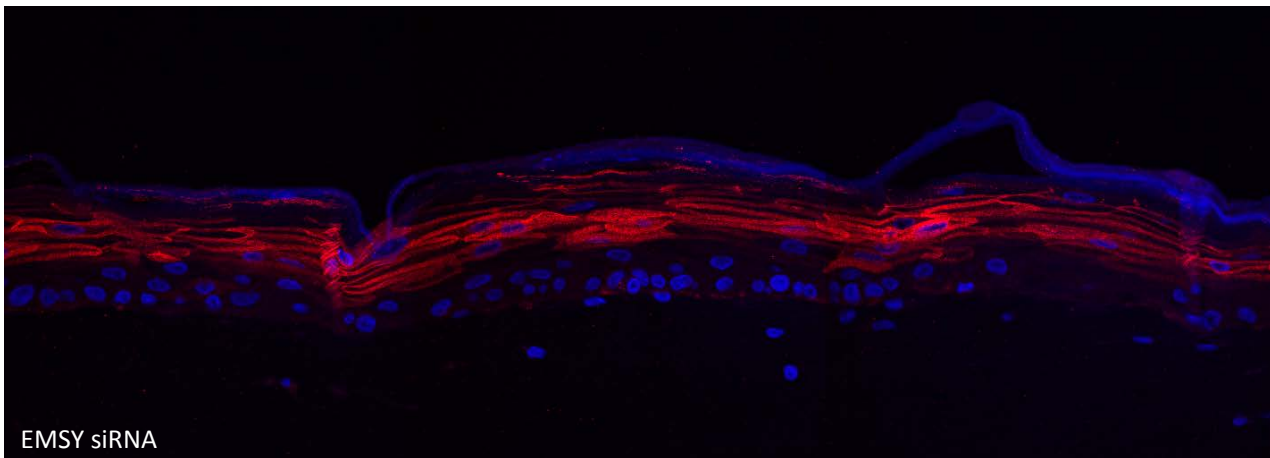
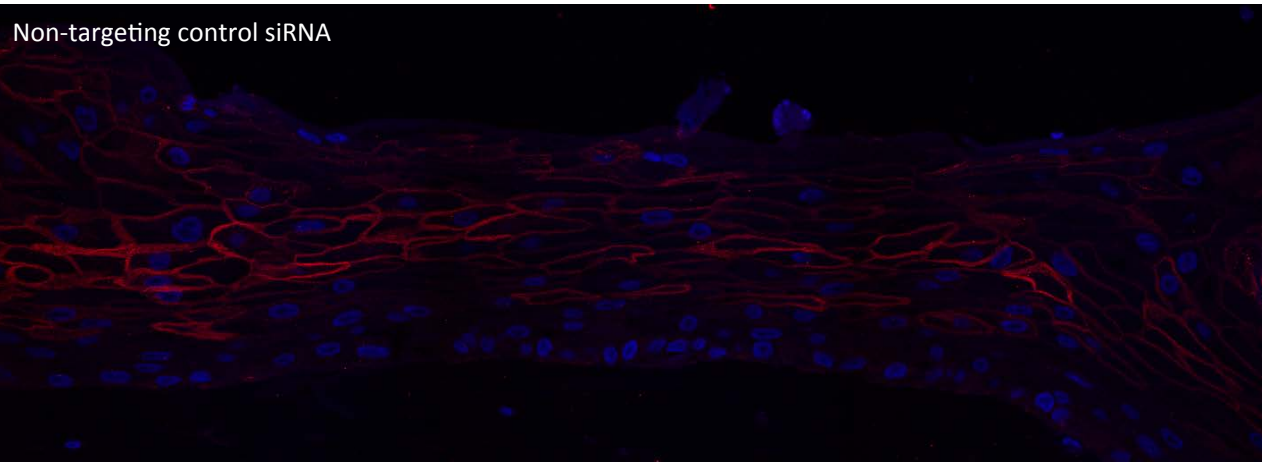


Corneodesmosin DAPI
Composite image created by Zeiss LSM710 microscope



Desmocollin DAPI

Composite image created by Zeiss LSM710 microscope



mTOR DAPI

Composite image created by Zeiss LSM710 microscope

