

Supplementary Online Content

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eTable 1. Histologic features examined

eTable 2. Custom PCR primers and probes

eTable 3. Fold induction of gene transcripts relevant to rosacea as determined by RT-PCR

This supplementary material has been provided by the authors to give readers additional information about their work.

eTable 1. Histologic features examined

Inflammation: Semi-quantitative evaluation using H&E-stained sections on glass slides (0 – 4 scale)

- 0—Normal. No inflammation
- 1—Minimal, focal inflammation limited to perivascular/perifollicular areas
- 2—Minimal-moderate inflammation not limited to perivascular/perifollicular areas
- 3—Moderate inflammation throughout dermis
- 4—Widespread inflammation

Connective tissue damage: Semi-quantitative evaluation using H&E-stained sections on glass slides (0 – 4 scale)

- 0—Normal. Intact collagen; essentially no elastosis
- 1—Minimal collagen fibril damage; thin, wispy collagen fibers with space between fibers, but no elastotic material
- 2—Elastotic material present but confined to area immediately beneath dermis; +/- collagen fibril damage
- 3—Elastosis throughout dermis; +/- collagen fibril damage
- 4—Widespread elastosis deep into the dermis accompanied by collagen fibril damage extending deep into dermis

Dilated vessels: Semi-quantitative evaluation using H&E stained sections on glass slides (0 – 4 scale)

- 0—Normal vessel caliber and number
- 1—One or two dilated vessels, but most vessels normal in size
- 2—More than 2 dilated vessels
- 3—Large number of vessels
- 4—Multiple vessels and/or extensive dilation

Sebaceous glands: Scanned sections evaluated morphometrically using Aperio slide scanning system

- 0—0% surface area covered by sebaceous glands
- 1—5%-10% surface area covered by sebaceous glands
- 2—10%--25% surface area covered by sebaceous glands
- 3—25%-50% surface area covered by sebaceous glands
- 4—>50% surface area covered by sebaceous glands

Additional Histologic Features:

Mast cells

- Cells counted in tissue sections after immunoperoxidase staining for mast cell tryptase.
- Mast cell degranulation assessed morphometrically; expressed as the percentage of surface area covered by immunostained (mast cell tryptase) material. Also assessed intensity of tryptase staining as a marker of degranulation.

Demodex mite - presence or absence detected in H&E stained sections on glass slides

Spongiosis - presence or absence detected in H&E stained sections on glass slides

Hyperkeratosis / parakeratosis - presence or absence in H&E stained sections

eTable 2. Custom PCR primers and probes

Oligonucleotide	Forward primer	Reverse primer
CXCR4	5'-GGTGGTCTATGTTGGCGTCT-3'	5'-TGGAGTGTGACAGCTTGGAG-3'
CGRP- α	5'-CATGTGGTTTGGTTCCTCTC-3'	5'-GGCCCTCATTTTCTGGTATT-3'
Substance P	5'-TCCAACATGAAAATCCTCGT-3'	5'-TTGCCCATTAATCCAAAGAA -3'
MMP-3	5'-TTCCTGATGTTGGTCACTTCAGA-3'	5'-TCCTGTATGTAAGGTGGGTTTTCC-3'
Decorin	5'-CCTGATGACCGCGACTTCGAG-3'	5'-TTTGGCACTTTGTCCAGACCC-3'
Type III collagen	5'-TCTTGGTCAGTCCTATGCGGATA-3'	5'-CATCGCAGAGAACGGATCCT-3'
Cathelicidin	5'-GTCACCAGAGGATTGTGACTTCAA-3'	5'-TTGAGGGTCACTGTCCCCATA-3'
DEFA-1	5'-CATGTTTTTCCTTGAGCCTGG-3	5'-CATGTTTTTCCTTGAGCCTGG-3'
Housekeeping gene 36B4	5'-ATGCAGCAGATCCGCATGT-3'	5'-TTGCGCATCATGGTGTTCTT-3'

eTable 3. Fold induction of gene transcripts relevant to rosacea as determined by RT-PCR

	AVERAGE FOLD CHANGE (p-value)			Significant 2-fold or greater induction
	ETR : Ctrl	TP : Ctrl	ETR:TP	Significant 2-fold or greater reduction
Neuropeptides				
CALCA	10.19 (<.001)	1.10 (.83)	9.30 (<.001)	Calcitonin-related peptide α ; encodes calcitonin-gene related peptide α (CGRP- α)
CALCB	6.99 (.05)	1.81 (.46)	3.86 (.13)	Calcitonin-related polypeptide β ; encodes calcitonin-gene related peptide β (CGRP- β)
TAC1	27.86 (<.001)	5.89 (.001)	4.73 (.002)	Tachykinin, precursor 1; encodes substance P, neurokinin A, neuropeptide K, neuropeptide γ
VIP	-1.67 (.02)	-1.02 (.93)	-1.64 (.02)	Vasoactive intestinal peptide
Matrix Remodeling				
COL1	4.68 (<.001)	2.10 (.04)	2.22 (.03)	Type I procollagen
COL3	5.19 (.001)	1.88 (.13)	2.76 (.02)	Type III procollagen
CYR61	3.89 (<.001)	1.51 (.19)	2.58 (.01)	Cysteine rich, angiogenic inducer, 61
DCN	10.02 (<.001)	1.32 (.49)	7.61 (<.001)	Decorin
MMP1	5.24 (.01)	4.93 (.01)	1.06 (.91)	Matrix metalloproteinase-1
MMP3	4.75 (.004)	1.08 (.87)	4.40 (.004)	Matrix metalloproteinase-3
MMP9	33.17 (<.001)	8.47 (.01)	3.92 (.07)	Matrix metalloproteinase-9
Innate Immunity				
CAMP	-1.57 (.21)	-1.41 (.21)	-1.11 (.63)	Cathelicidin antimicrobial peptide
DEFA1	92.27 (.001)	1.01 (.99)	91.51 (.001)	Defensin, α 1
HBD2	1.75 (.51)	20.62 (.01)	-11.80 (.01)	Human β defensin-2
HBD3	-1.61 (.39)	6.31 (.01)	-10.14 (.001)	Human β defensin-3
SCCE	-1.58 (.12)	2.15 (.02)	-3.39 (.001)	Stratum corneum chymotryptic enzyme (synonym Kallikrein-related peptidase 7 [KLK7])
SCTE	1.41 (.15)	2.07 (.02)	-1.47 (.15)	Stratum corneum tryptic enzyme (synonym Kallikrein-related peptidase 5 [KLK5])

	AVERAGE FOLD CHANGE (p-value)			Significant 2-fold or greater induction
	ETR : Ctrl	TP : Ctrl	ETR:TP	Significant 2-fold or greater reduction
Mast Cell Chemotaxis				
CXCL12	2.52 (.002)	1.70 (.06)	1.49 (.10)	chemokine (C-X-C motif)
CXCR4	8.20 (<.001)	3.20 (.03)	2.56 (.04)	chemokine (C-X-C motif)
Other Inflammatory Markers				
CCL20	-1.35 (.52)	1.46 (.52)	-1.97 (.30)	Chemokine (C-C motif)
CCL27	1.04 (.96)	-2.13 (.45)	2.21 (.45)	Chemokine (C-C motif)
CTGF	-1.57 (.17)	1.05 (.84)	-1.65 (.15)	Connective tissue growth factor
CXCL-1	1.07 (.92)	2.32 (.30)	-2.16 (.30)	chemokine (C-X-C motif)
CXCL14	1.28 (.25)	1.67 (.04)	-1.30 (.25)	chemokine (C-X-C motif)
FCERIB	-25.34 (.01)	-2.38 (.23)	-10.67 (.03)	Fc fragment of IgE, high affinity I, receptor for; β polypeptide
IL 1B	1.14 (.95)	-1.02 (.95)	1.16 (.95)	Interleukin-1, β
IL-8	2.07 (.47)	1.62 (.47)	1.28 (.58)	Interleukin 8
IL-12A	-1.08 (.87)	1.37 (.76)	-1.48 (.76)	Interleukin 12A
IL-12B	2.66 (.01)	2.84 (.01)	-1.07 (.82)	Interleukin 12B
IL-17A	1.41 (.55)	2.17 (.51)	-1.54 (.55)	Interleukin 17A
IL-17F	1.90 (.43)	1.58 (.43)	1.20 (.63)	Interleukin 17F
IL-23A	-1.70 (.32)	1.31 (.50)	-2.24 (.11)	Interleukin 23A
IL-31	3.98 (.24)	1.42 (.65)	2.80 (.24)	Interleukin 31
TNFA	2.09 (.02)	-1.41 (.26)	2.96 (.002)	Tumor necrosis factor α
TPSAB1	1.37 (.62)	-1.10 (.81)	1.50 (.62)	Tryptase-1