• •				Size (d.nm):	% Intensity:	St Dev (d.nm):
A)	Z-Average (d.nm):	224.9	Peak 1:	265.8	100.0	113.9
0.000	Pdi:	0.163	Peak 2:	0.000	0.0	0.000
	Intercept:	0.968	Peak 3:	0.000	0.0	0.000
	Result quality :	Good				



				Mean (mV)	Area (%)	St Dev (mV)
B)	Zeta Potential (mV):	33.6	Peak 1:	33.7	100.0	5.91
	Zeta Deviation (mV):	5.57	Peak 2:	0.00	0.0	0.00
	Conductivity (mS/cm):	2.10	Peak 3:	0.00	0.0	0.00





Supplementary Figure 1





Supplementary Figure 2

Supplementary Figure 3



Supplementary Figure 4



Mice divided into 4 groups: IMQ(-), IMQ(+), IMQ+EGCG and IMQ+nano-EGCG



skin 20x DAPI 488 texas red.csl





Supplementary Figure 6



Supplementary Figure 7

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Supplementary Figure 8A



## Supplementary Figure 8B

## SUPPLEMENTARY MATERIALS AND METHODS

# Chitosan-based nanoformulated epigallocatechin-3-gallate EGCG modulates keratinocyte-induced responses and alleviates imiquimod-induced murine psoriasiform dermatitis

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### **Supplementary Materials and Methods**

#### Transmission electron microscopy analysis of nanoparticle uptake into cells

**and tissues.** Processing and documentation of samples were as previously described<sup>1</sup> with some modifications. Briefly, tissues and cultured cells samples were fixed as follows: The skin tissue and cultured cell samples (on entire glass coverslips) were fixed in 2.5% glutaraldehyde, 2.0% paraformaldehyde in 0.1M sodium phosphate buffer (PB), pH 7.4 for 2 hrs @ room temperature (RT) followed by 5 x 5 minute rinses in 0.1M PB. Samples were then post-fixed in 1% Osmium Tetroxide (OsO4), 1% potassium ferrocyanide in PB for 1 hr @ RT, rinsed in PB as before, followed by 3 x 5 minute final rinses in distilled water to clear the phosphate. Skin sections and cultured cells were dehydrated in a graded ethanol series (35, 50, 70, 80, 90% for 10 minutes each step, 95% for 20 minutes, and finally 100% for 3 x 10 minutes) at RT and then transitioned in propylene oxide (PO) 2 x 7 minutes at RT. For tissue sections, fully dehydrated samples were infiltrated with increasing concentrations of PolyBed 812 (Polysciences Inc. Warrington, PA) and PO mixtures in the following order: 75% PO: 25% Polybed 812 for 1 hour at RT, 50% PO: 50% Polybed 812 for 1 hour at RT, 25% PO: 75% Polybed 812 overnight @ RT, followed by a final incubation in 100% Polybed 812 4 x 1 hour at 60°C in a drying oven. Embedding and polymerization took place in fresh PolyBed 812 for 24 hours at 60°C.

For infiltration and embedding of cultured cells, Durcupan ACM (Fluka AG, Switzerland) resin was used. Increasing concentrations of accelerated Durcupan (10ml A/M, 10ml B, 300ul C, 100ul D components) were used for infiltration. All infiltration steps were done with the coverslips, cell side up, in aluminum weighing dishes. The weighing dishes were placed in covered glass petri dishes to minimize PO evaporation in the following order. 75% PO : 25% Durcupan for 1 hour at RT, 50% PO: 50% Durcupan for 1 hour at RT, 25% PO : 75% Durcupan overnight @ RT and 100% Durcupan for 3 x 1 hour at 60°C in a drying oven. The cultured cells were then embedded in open aluminum weighing dishes @ 60°C in a drying oven overnight until polymerized. Following polymerization, the samples were cooled, and the aluminum dishes stripped off. The excess epoxy resin was scraped off the bottom of the embedded coverslips to expose the glass surface. The samples were then floated in concentrated hydrofluoric acid, glass side down, for 15 minutes to etch off the glass, revealing the embedded cell culture, and mounted on stubs for sectioning on a Leica EM UC6 ultramicrotome at 100nm. The sections were collected on 2x1 slot

Pioloform/carbon coated Cu grids (EMS Hatfield, PA) for tissue samples and 300 mesh thin bar Cu grids (EMS Hatfield, PA) for cell culture samples. Post-staining was performed with uranyl acetate (UA) and lead citrate. The sectioned samples were viewed at 80kV on a Philips CM120 transmission electron microscope, equipped with MegaView III camera (Olympus Soft Imaging System Lakewood, CO).

#### Keratinocyte culture, and determination of cell viability and proliferation by

MTT and Trypan Blue Exclusion assays. Each vial of frozen primary Normal human epidermal keratinocytes (NHEKs) was thawed and cultured in CellnTec progenitor cell culture (Cn-PR) or EpiLife medium (with added HKGS), all supplemented medium with antibiotics/antimycotics and were maintained for about 2 months (8 passages). Cells were cultured under standard cell culture conditions at 5% CO<sub>2</sub> and 37 °C in a humidified atmosphere incubator<sup>2</sup>. The comparative effects of voided-chitosan-nanoparticles (CHIT-Void-NPs), free EGCG (native EGCG) and nanoEGCG (CHIT-nanoEGCG) on NHEK viability was determined using a 3- (4,5dimethylthiazol-2yl)-2,5-diphenyltetrazolium bromide (MTT) assay (Sigma-Aldrich, St. Louis, MO). NHEKs were seeded at a density of  $2 \times 10^4$  cells/well in 24-well poly(D)lysine (0.1 mg/mL; Sigma-Aldrich, St. Louis, MO) pre-coated plates in 1 ml culture medium and incubated at 37 °C and 5% CO2. At 80% confluence, cells were treated in the presence or absence of voided nanoparticles (vehicle control), increasing concentrations of free EGCG (0, 5, 10, 20, 40, 80 µM) or nanoEGCG (concentration equivalents of active drug encapsulated in chitosan nanoparticle were 0, 1.25, 2.5, 5, 10, and 20 µM) in culture medium. All treatment formulations (freeEGCG, nanoEGCG and voided-chitosan-nanoparticles) were suspended in PBS, and then suitably diluted in respective culture media. After 24-48 h of incubation, media were removed and cells were rinsed with PBS and incubated for 3 h with 300µL of MTT solution (0.5 mg/ml; Sigma-Aldrich). The MTT solution was removed and the formazan crystals were solubilized in DMSO (300µL) under slow shaking. Absorbance for each sample were spectrophotometrically recorded at 570 nm on a BioTek microplate reader (Bio-TEK Instruments, Inc., Winooski, VT). Experiments were repeated three times, each in quadruplicate, with similar results. The comparative effects of freeEGCG, nanoEGCG and voided-chitosan-nanoparticles were assessed as percentage of cell viability (expressed as average absorbance at 570nm), relative to the viability of untreated controls, which was set at 100%. IC 50 values were calculated for free EGCG and nanoEGCG.

For cell viability assays, the Trypan Blue Dye Exclusion (TBDE) method was employed as described previously<sup>3</sup>. Briefly, NHEKs were seeded overnight at  $3x10^5$  cells/100mm petri dish and control plates (t=0) were harvested and cell counted. Growth media were replenished with fresh corresponding complete medium supplemented with or without free EGCG (10 and 20  $\mu$ M) or nanoEGCG (5-10  $\mu$ M), and further incubated for 48 h. The cells were then harvested using 0.025% trypsin containing 1mM EDTA, stained with trypan blue solution and counted in a BioRad TC10 cell counting chamber (Bio-Rad Laboratories Inc., Des Plaines, IL). Trypsinized cells were evaluated for total viable cell number harvested (after 48h) in relation to the counts obtained for control (t=0) plates prior to treatment (0h). Cultures treated with voided-chitosan-nanopartic les (controls) were considered as 100% viable. All experiments were performed three times, each in quadruplicate, and data are represented as means  $\pm$  SD.

For recombinant human (rh) interleuk in (IL)-22 stimulation, near-confluent NHEK cultures were pre-treated with or without different concentrations of free EGCG (20  $\mu$ M) or nanoEGCG (equivalent bound concentrations of EGCG (5 and 10  $\mu$ M) for 6 h. Cells were then co-treated with or without rhIL-22 (20 ng/mL) for additional 42 h (for a total of 48 h exposure to the drugs), harvested and analyzed for cell viability by MTT assay.

12-O-tetradecanoyl-phorbol-13-acetate (TPA) induction of inflammatory responses and cytokines: For TPA stimulation, near-confluent NHEK <u>cultures</u> were pretreated with or without free EGCG (20  $\mu$ M) or nanoEGCG (equivalent bound EGCG concentration of 5  $\mu$ M) for 18 h, followed by co-treatment with or without TPA (100 ng/mL) for the last 6 h (making a total of 24 h of exposure to the drugs). The conditioned media were collected, centrifuged and supernatants were stored at -80°C, and were subjected to a single freeze-thaw cycle prior to use in the Procarta multiplex cytokine immunoassay. The concentrations of secreted cytokines (IL-1 $\beta$ , IL-6, IL-8, IL-10, IFN- $\gamma$ , TGF- $\alpha$  and CXCL2) in conditioned media was measured using a mix-matched 6-Plex Procarta<sup>TM</sup> immunoassay (Affymetric, eBiosciences) following the manufacturers protocol as detailed below.

Cell lysate preparation from cultured cells or skin tissue and immunoblotting analysis: NHEK cultures were pre-treated with or without voided-chitosan-nanoparticles, free EGCG (0-20  $\mu$ M) or nanoEGCG (equivalent bound EGCG concentrations of 0-5  $\mu$ M) for a total of 48 h prior to harvest. Cells were then washed with cold PBS (10 mM sodium phosphate, pH

7.4) and incubated in 1X lysis buffer (50 mM Tris–HCl, 150 mM NaCl, 1 mM ethyleneglycolbis(aminoethylether)-tetraacetic acid, 20 mM NaF, 100 mM Na<sub>3</sub>VO<sub>4</sub>, 0.5% NP-40, 1% Triton X-100, 1 mM phenylmethylsulfonyl fluoride, pH 7.4), with freshly added protease inhibitor cocktail Set III (Calbiochem, La Jolla, CA) on ice for 30 min. Cells were scraped, collected and transferred into microfuge tubes, and suspended cell aggregates was sheared by passing several times through 22.5-gauge syringe needles to break up cell <u>and incubated on ice for 20 minutes</u>. The lysates were cleared by centrifugation at 14000xg for 30 min at 4°C, and the protein concentrations were determined using the BCA protein assay kit according to the manufacturer's protocol. Lysates were aliquoted and frozen at -80°C for further analysis. Aliquots containing 20 µg total protein per sample were subjected to SDS-PAGE followed by immunoblotting, chemiluminescence detection and analysis as earlier described<sup>4,5</sup>. Equal loading of protein was confirmed by stripping and reprobing <u>the membrane blots</u> for  $\beta$ -actin. Results displayed in figures are representative of three independent experiments.

**Toluidine blue staining:** For quantification of skin mast cell numbers, skin tissue sections were stained with toluidine blue solution (Sigma-Aldrich, St. Louis, MO) for 2-3 min, dehydrated in alcohol, cleared in xylene and mounted using a xylene based permount. The total numbers of mast cells, and the numbers of degranulated mast cells per field of view, were determined under a light microscope at 20x and 40x magnifications.

**ProcartaPlex<sup>TM</sup> Multiplex Bead-based Immunoassays for Cytokines and Chemokines:** Commercially available human and mouse Procarta Multiplex Bead Immunoassay kits (Affymetrix/eBioscience, Santa Clara, CA), were used to determine cytokine and chemokine levels in conditioned media from TPA-stimulated NHEK cells, as well as in mouse skin lysates as earlier described<sup>6</sup>. These assays included a wide range of cytokines and chemokines that reflect key processes and responses relating to the activation of inflammation. Analyses with the 36-Plex Mouse Procarta assay allowed us to monitor changes in the expression of immune factors in the IMQ-induced psoriasis-like skin inflammation model in mice, and to determine the effectiveness of free EGCG and nanoEGCG interventions. In addition, we utilized a customized mix-matched human procarta 6-Plex pro-inflammatory cytokine panel to investigate the role of free EGCG and nanoEGCG in modulating the secretion of pro-inflammatory cytokines from TPAinduced NHEKs as previously described<sup>6</sup>. Briefly, beads of defined spectral properties conjugated to analyte-specific capture antibodies were combined with test samples (25 μL for 36-Plex mouse

tissue lysates or 50 µL for Human 6-Plex NHEK cultured supernatants) in separate wells of a 96well black side/transparent bottom filter microplate. These were then incubated at RT for 120 minutes and further processed following the manufacturer's instructions, and as previously described<sup>6</sup>. All skin lysates and cultured supernatants were analyzed in triplicate along with serial standards (7-point dilutions). Analytes were allowed to bind to each of the capture antibodies on the beads, several washes were performed to remove non-specifically bound proteins, and analytespecific biotinylated detection antibodies were added and incubated with the beads at RT for 30-60 minutes. During this incubation, the analyte-specific biotinylated detection antibodies bind to specific epitopes on the immobilized analytes. Following the binding, incubation and washes, streptavidin conjugated to Phycoerythrin, (SA-PE or Streptavidin-PE), a pigment complex that serves as a fluorescent tag was added and samples were incubated at RT with slow shaking for 30 minutes. During this final incubation, Streptavidin-RPE binds to the biotinylated detector antibodies associated with the immune complexes on the beads, forming a four-member solid phase sandwich. After washing cycles to remove unbound SA-PE, excess reading buffer was added and incubated at room temperature for a minimum of 5 minutes in the dark with shaking and stored at 4° overnight. Fluorescently tagged beads were analyzed with the xMAP Luminex reader (Luminex instrument, Austin, TX) for quantitative analysis. The spectral properties of the beads and the amounts of associated PE fluorescence were monitored to determine the concentration of the following analytes; i) for mouse skin lysates: interleukin (IL)-1 family members IL-1 $\alpha$ , IL-1 $\beta$ and IL-18; IL/ $\gamma$  chain family members IL-2, IL-4 and IL-15/IL-15R; IL/ $\beta$  chain family members IL-3, IL-5 and GM-CSF (CSF2); IL-6 (gp130) family members IL-6, IL-31 and LIF; IL-12p70; IL-10; IL-9; IL-17A; IL-22; IL-23; IL-27; IL-13; IFNa2 (IFNa); IFNy; IL-28; TNFa; CC chemokines CCL2 (MCP-1), CCL-3 [macrophage inflammatory protein-1a (MIP-1a)], CCL4 (MIP-1ß), CCL5 [also known as regulated on activation, normal T cell expressed and secreted (RANTES)], CCL7 (MCP-3), CCL-11 (eotaxin); CXC chemokines - CXCL-1 [growth-regulated oncogene  $\alpha$  (GRO $\alpha$ )], CXCL-2 (MIP-2), CXCL-5 [epithelial-derived neutrophil-activating peptide 78 (ENA-78)], CXCL-10 (IP-10); and growth/cellular factors-M-CSF (CSF1) and G-CSF (granulocyte-colony stimulating factor-3, also called CSF3). For culture supernatants obtained from human NHEKs: IL-1β, IL-6, CXCL8 (IL-8), TNFa, TGF-a, and CXCL2. The cytokine and chemokine concentrations were determined by importing data into Procarta Plex<sup>TM</sup> Multiplex

Analyst software v.1.0 (Affymetrix/eBioscience, Santa Clara, CA) for analysis as earlier described<sup>6</sup>.

Antibody/Antigen	Species	Company	Cat. no	Dilutions	
	I	1	L	Western blot	immunostaining (IHC/IF)
CD31	Rabbit	Abcam <sup>1</sup>	Ab28364	n.d.	1:250
CD4 <sup>+</sup>	Rabbit	Cell Marque <sup>2</sup>	104R-14	n.d.	1:300
caspase-14	Rabbit Po.	Imgenex, Novus Biologicals, LLC3	IMG-5713	1:1000	1:1000/100
Filaggrin	Mouse Mo.	Novocastra Laboratories Ltd <sup>4</sup>	15C10	1:500	1:200
Jun B	Rabbit	Santa Cruz <sup>5</sup>	Sc-46X	n.d.	1:200
Involucrin	Mouse Mo.	NeoMarker <sup>6</sup>	SY5	1:1000	n.d.
Keratin-1	Rabbit Po.	Covance <sup>7</sup>	PRB-	1:1000	1:100
Keratin-10	Rabbit Po.	Covance <sup>7</sup>	PRB-159P	1:1000	1:1000 and 1:500
Loricrin	Rabbit	Santa Cruz <sup>5</sup>	Sc-133757	n.d.	1:500
transglutaminase- 1(TGase-1)	Mouse Mo.	Biomed. Technol. Inc. <sup>8</sup>	BT-621	1:500	n.d.
HRP donkey anti- goat IgG		Santa Cruz Biotech <sup>5</sup>	Sc-2033	1:5000	n.d.
β-actin	Mouse	Sigma Aldrich <sup>2</sup>	A5441	1:5000	n.d.
Alexa Fluor 488	Rabbit	Invitrogen/ Thermo Fisher Scientific <sup>9</sup>			1: 600
Alexa Fluor 594	Mouse	Invitrogen			1:600

## Supplementary Table : Details of antibodies used in this study and their sources.

**Company & addresses:** <sup>1</sup>Cambridge, MA, USA; <sup>2</sup>St. Louis, MO, USA; <sup>3</sup>Littleton, CO, USA; <sup>4</sup>Leica Biosystems Inc. Buffalo Grove, IL, USA; <sup>5</sup>Santa Cruz, CA, USA); <sup>6</sup>Fremont, CA, USA; <sup>7</sup>Madison, WI, USA; <sup>8</sup>Mount Arlington, NJ, USA; <sup>9</sup>Thermo Fisher Scientific Rockford, IL, USA; n.d. not determined.

## **Supplementary Figure Legends**

Supplementary Figure - 1. (A) Size measurement of Voided Chitosan-nanoparticles (CHI-TPP-Void-NPs) using Dynamic Light Scattering (DLS), and (B) Zeta Potential Measurement.
Supplementary Figure - 2: Transmission Electron Microscope photomicrograph showing the size and morphology of Voided Chitosan-nanoparticles (CHI-TPP-Void-NPs).
Supplementary Figure - 3: *In vitro* release kinetic profiles of EGCG from nanoEGCG in

phosphate buffered saline (pH 7.4) at 37 °C compared to that of free EGCG.

<u>Supplementary</u> Figure – 4. Gross morphological changes in cells treated with nanoEGCG and free EGCG. Near-confluent NHEKs were pretreated for 48 h without (control) or with EGCG (20  $\mu$ M or 40  $\mu$ M) or with nanoEGCG (5 or 10  $\mu$ M) as indicated in panels A-F, prior to taking phase contrast photomicrographs (Magnification x200), Bar = 50  $\mu$ m. Panels G and H were treated with a 20  $\mu$ M equivalent dose of nanoEGCG; at this high concentration, nanoparticles floated on cell surface, blurring the micrographic images.

<u>Supplementary</u> Figure – 5. Schematic illustration of the experimental protocol before and after IMQ-induced psoriasis-like skin inflammation in Balb/c mice, including topical application of nanoEGCG or free EGCG. Briefly, 6–8-week-old mice were shaven (+Nair), then topically treated starting on day 3 for 14 consecutive days with control Vaseline ointment (Group 1, IMQ(-)) or Vaseline containing 5% IMQ-containing cream (Aldara 3.125mg/cm<sup>2</sup> of skin-area, and 1.63 mg/ ear/day) for Groups 2, 3 and 4. Starting on day 9, Group 3 received IMQ-cream and (free) EGCG (1 mg/cm<sup>2</sup>/day, denoted "IMQ+EGCG), while Group 4 received nanoEGCG (48 µg/cm<sup>2</sup> /day, denoted IMQ+nanoEGCG) for a further 9 days. Mice were sacrificed on day 17 and samples were taken for further analyses. It is worth noting that in preliminary studies conducted, data for voided chitosan-nanoparticles (CHI-TPP-Void-NPs) were similar to control Vaseline treatment group. Because we are also comparing the effects of free EGCG to that of nanoEGCG, we decided to present only vaseline treated control data.

Supplementary Figure – 6. Schematic filter libraries for the three different chromogens (DAPI, Alexa488 (488), and Texas Red) generated using the Nuance imaging algorithm software. A spectral library of 3 chromogens (DAPI, Alexa488, and Texas Red) was built using the Nuance software with more than 95% accuracy.

Supplementary Figure – 7. Representative bar graphs showing expression of inflammatory mediators that are not significantly altered by IMQ, EGCG or nanoEGCG treatments.

Supplementary Figure – 8. Effect of topical application of free EGCG and nanoEGCG on the expression of inflammatory cytokines/chemokines in IMQ-treated mice. Bar graphs show (A) protein expression of cytokines that are pro-inflammatory and anti-inflammatory, Th1, Th2 and activators of granulocytes and/or monocytes/macrophages, and (B) protein expression of Th17/Th22/Treg cytokines and type-1/2 and granulocytes and/or monocytes/macrophage chemokines. Immunoassays of proinflammatory mediators expressions were performed on skin lysates from the four treatment groups. Briefly, back skin was collected from mice, and cell lysates

were prepared (see Supplementary Methods) and a Procarta 36-multiplex immunoassay was performed to evaluate the expression levels of cytokines and chemokines in samples from the control (IMQ-), IMQ+, IMQ+EGCG and IMQ+nanoEGCG groups. (A) Expression of Th1/Th2 cytokines and activators of granulocytes and/or monocytes/macrophages (B) Expression of Th17/Th22/Treg cytokines, type-1/type-2 chemokines, and granulocytes and/or monocyte/macrophage chemokines. Data analyses for all samples are presented and comparisons between groups that meet significance criteria in post hoc analyses are indicated by lines above groups being compared and asterisks denoting P values from two-sample t-test comparisons as follows: \* p < 0.05; \*\* p < 0.01; \*\*\* p < 0.001; and \*\*\*\* p < 0.0001.

#### References

A. M. Schrand, J. J. Schlager, L. Dai and S. M. Hussain, Preparation of cells for assessing ultrastructural localization of nanoparticles with transmission electron microscopy. *Nat Protoc*. 2010;5:744-57

J. C. Chamcheu, H. C. Pal, I. A. Siddiqui, V. M. Adhami, S. Ayehunie, B. T. Boylan, et al., Prodifferentiation, anti-inflammatory and antiproliferative effects of delphinidin, a dietary anthocyanidin, in a full-thickness three-dimensional reconstituted human skin model of psoriasis. *Skin Pharmacol Physiol.* 2015;28:177-88

J. C. Chamcheu, F. Afaq, D. N. Syed, I. A. Siddiqui, V. M. Adhami, N. Khan, et al., Delphinid in, a dietary antioxidant, induces human epidermal keratinocyte differentiation but not apoptosis: studies in submerged and three-dimensional epidermal equivalent models. *Exp Dermatol.* 2013;22:342-8

J. C. Chamcheu, M. I. Chaves-Rodriquez, V. M. Adhami, I. A. Siddiqui, G. S. Wood, B. J. Longley, et al., Upregulation of PI3K/AKT/mTOR, FABP5 and PPARbeta/delta in Human Psoriasis and Imiquimod-induced Murine Psoriasiform Dermatitis Model. *Acta Dermato-venereologica*. 2016

J. C. Chamcheu, H. Navsaria, I. Pihl-Lundin, M. Liovic, A. Vahlquist and H. Torma, Chemical chaperones protect epidermolysis bullosa simplex keratinocytes from heat stress-induced keratin aggregation: involvement of heat shock proteins and MAP kinases. *J Invest Dermatol.* 2011;131:1684-91

J. C. Chamcheu, V. M. Adhami, S. Esnault, M. Sechi, I. A. Siddiqui, K. A. Satyshur, et al., Dual Inhibition of PI3K/Akt and mTOR by the Dietary Antioxidant, Delphinidin, Ameliorates Psoriatic Features In Vitro and in an Imiquimod-Induced Psoriasis-Like Disease in Mice. *Antioxid Redox Signal*. 2016