Supplemental Material and Methods

Cell culture

HaCaT keratinocytes (Human adult low Calcium high Temperature) were grown to confluence in a humidified atmosphere containing 5%CO₂ at 37°C in Dulbecco’s modified Eagle’s medium (DMEM) (Life Technologies; Carlsbad; CA; USA) supplemented with 10% fetal calf serum (Biochrom, Berlin, Germany), 50units/ml penicillin (AppliChem, Darmstadt, Germany), 50μg/ml streptomycin (AppliChem) and an adjusted Ca²⁺- concentration of 1.8mM. Confluent monolayers were used for Dispase-based keratinocyte dissociation assay.

Dispase-based keratinocyte dissociation assay

Konfluent HaCaT monolayers were washed once with pre-warmed HBSS (Sigma Aldrich; Munich, Germany) and incubated with HBSS containing Dispase II (>2,4U/ml; Sigma Aldrich) for 30min at 37°C to remove monolayers from well bottom. Afterwards Dispase solution was replaced by HBSS to stop reaction and a defined sheer stress was applied by pipetting the monolayer 5 times with a 1ml electronic pipet. Numbers of fragments were counted and represent a measure for loss of intercellular adhesion.

MTT-assay

To check viability of conjunctiva samples, 12h cultured samples were washed once with PBS and incubated with Thiazolyl Blue Tetrazolium Bromide (MTT) (Sigma Aldrich, Munich, Germany) at a final concentration of 0.5mg/ml for 20min. Viable probes show a colour change to blue-violett, while dead tissue remains unstained. Some probes were heated to 65°C for 15min before MTT-assay and were used as a negative control.