



(51) International Patent Classification:

A61Q 5/02 (2006.01) A61K 8/49 (2006.01)

A61Q 5/12 (2006.01)

(21) International Application Number:

PCT/EP20 14/056082

(22) International Filing Date:

26 March 2014 (26.03.2014)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

13 16675 1.1 7 May 2013 (07.05.2013) EP

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(81) **Designated States** (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) **Designated States** (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:

- as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(H))
- as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(in))
- of inventorship (Rule 4.17(iv))

Published:

- with international search report (Art. 21(3))

(54) **Title:** USE OF CLIMBAZOLE

(57) **Abstract:** The invention relates to the cosmetic use of climbazole to provide an epidermal skin barrier benefit, and to a cosmetic method to provide an epidermal skin barrier benefit by applying to the skin a composition comprising climbazole.



WO 2014/180600 A1

Use of Climbazole

The invention relates to the use of climbazole to provide an epidermal skin barrier benefit.

5 Climbazole is known as an anti-fungal agent and as such is used as an ingredient in anti-dandruff shampoos.

EP 2 628 479 discloses cosmetic, non-therapeutic use of climbazole for influencing the natural production of components of the extracellular matrix in the dermis, e.g. collagen,
10 elastin, and glycosaminoglycan.

EP 2 633 887 discloses a cosmetic composition comprising climbazole, tocopherol, glycyrrhizic acid and hyaluronic acid. In addition, the cosmetic, non-therapeutic use of the composition is disclosed for the reduction of wrinkles and fine lines or for the
15 treatment of the signs of tired and sagging skin and to improve skin hydration.

None of the aforementioned documents discloses the effect of climbazole on the epidermal skin barrier, or the effect of climbazole on the production of proteins in the epidermis. It is well known (Marks, James G; Miller, Jeffery (2006)., Lookingbill and
20 Marks' Principles of Dermatology (4th ed.), Elsevier. pp. 1-7) that the dermis and epidermis are distinct and separate parts of the skin and that the epidermis is the outer part of the skin which and acts as the body's major barrier against the environment.

Summary of the Invention

25 The epidermal skin barrier is the natural protection layer of the body. The epidermal skin barrier forms an effective two-way protection barrier from the environment, preventing unwanted invasion of chemicals from outside, and preventing unregulated loss of water and nutrients from inside. We have found that climbazole can provide benefits to the epidermal skin barrier.

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The invention thus provides in a first aspect the cosmetic use of climbazole to provide an epidermal skin barrier benefit.

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A second aspect of the invention relates to a cosmetic method for provision of an epidermal skin barrier benefit, comprising applying to the skin a composition comprising climbazole.

- 5 Preferably in the use or method, the epidermal skin barrier benefit is selected from: strengthening the skin barrier and/or repair of the skin barrier.

Preferably in the use or method, the epidermal skin barrier benefit is a scalp skin barrier benefit.

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Preferably in the use or method, the climbazole is used in a personal care formulation comprising climbazole at a level of from 0.1 to 5 wt.%, more preferably from 0.1 to 2.5 wt.%.

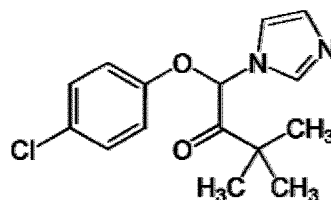
- 15 Preferably in the use or method, the personal care formulation is a hair and/or scalp treatment composition. More preferably the hair and/or scalp treatment composition is a shampoo, conditioner or a leave-on composition.

Detailed Description of the Invention

- 20 Climbazole is an anti-fungal agent, used in anti-dandruff formulations. It is believed to act as an anti-fungal against *Malassezia* which is associated with dandruff symptoms.

Climbazole has the following chemical structure:

25



- 30 The epidermal skin barrier benefit of climbazole was ascertained by microarray analysis. Human keratinocyte skin cells were treated with various doses of climbazole for a certain amount of time. Cellular mRNA (transcriptome) was extracted, and microarray analysis performed. This analysis highlights activity of 20,000 genes and illustrates changes in transcriptome due to climbazole treatment.

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Analysis of the changes due to the effects of the climbazole was carried out to find gene expression changes due to treatment by climbazole. A group of genes that showed a large transcriptome change was a group relating to epidermal differentiation. These genes encode proteins which then have biological functions. The specific epidermal differentiation genes that were positively affected by treatment with climbazole are associated with the skin and specifically the skin barrier. Upregulation of these genes thus positively benefits the epidermal skin barrier, and specifically strengthening the skin barrier and/or repair of the skin barrier.

Formulations

The climbazole is preferably used in a personal care formulation to deliver the epidermal skin barrier benefit. The personal care composition of the invention may be in any form but is preferably suitable for topical application to the skin either as a leave-on or rinse-off composition.

More preferably, the personal care formulation is a hair and/or scalp treatment composition. Preferably the hair and/or scalp treatment composition is a shampoo, conditioner or a leave-on composition.

Preferably the personal care composition comprises a cationic deposition polymer.

Suitable cationic deposition polymers may be a homopolymer or be formed from two or more types of monomers. The weight-average molecular weight of the polymer will generally be between 5 000 and 10 000 000 unified atomic mass units, typically at least 10 000 and preferably from 100 000 to 2 000 000. The polymer will have cationic nitrogen containing groups such as quaternary ammonium or protonated amino groups, or a mixture thereof.

Preferably, the cationic deposition polymer is cationic polysaccharide. Such cationic polysaccharide includes for example, cationic celluloses and hydroxyethylcellulose, cationic starches and hydroxyalkyl starches, cationic polymer based on guar gum. More preferably the cationic deposition polymer is (or comprises) cationic polygalactomannan,

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especially guar or cassia derived polygalactomannan modified with hydroxypropyl trimonium chloride.

Specific non-limiting examples of cationic guar polymer includes Jaguar® C-14S,
 5 Jaguar® C-13S, Jaguar® C-17, Jaguar® C-500, Jaguar® C-162, Jaguar® Excel.

When a cationic polymer is present, it is preferred that compositions contain from 0.01% to 2% wt. of the composition cationic deposition polymer, more preferably from 0.05 to 0.5% wt. and most preferably from 0.08 to 0.25% by weight of the composition .

10

Preferably the personal care composition comprises cleansing surfactant.

Examples of suitable anionic cleansing surfactants are the alkyl sulphates, alkyl ether sulphates, alkaryl sulphonates, alkanoyl isethionates, alkyl succinates, alkyl
 15 sulphosuccinates, alkyl ether sulphosuccinates, N-alkyl sarcosinates, alkyl phosphates, alkyl ether phosphates, and alkyl ether carboxylic acids and salts thereof, especially their sodium, magnesium, ammonium and mono-, di- and triethanolamine salts. The alkyl and acyl groups generally contain from 8 to 18, preferably from 10 to 16 carbon atoms and may be unsaturated . The alkyl ether sulphates, alkyl ether sulphosuccinates, alkyl ether
 20 phosphates and alkyl ether carboxylic acids and salts thereof may contain from 1 to 20 ethylene oxide or propylene oxide units per molecule.

Typical anionic cleansing surfactants for use in compositions include sodium oleyl succinate, ammonium lauryl sulphosuccinate, sodium lauryl sulphate, sodium lauryl ether
 25 sulphate, sodium lauryl ether sulphosuccinate, ammonium lauryl sulphate, ammonium lauryl ether sulphate, sodium dodecylbenzene sulphonate, triethanolamine dodecylbenzene sulphonate, sodium cocoyl isethionate, sodium lauryl isethionate, lauryl ether carboxylic acid and sodium N-lauryl sarcosinate.

30 Preferred anionic surfactants are the alkyl sulfates and alkyl ether sulfates. These materials have the respective formulae R_2OSO_3M and $R-iO(C_2H_4O)_xSO_3M$, wherein R_2 is alkyl or alkenyl of from 8 to 18 carbon atoms, x is an integer having a value of from about 1 to about 10, and M is a cation such as ammonium, alkanolamines, such as triethanolamine, monovalent metals, such as sodium and potassium, and polyvalent

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metal cations, such as magnesium, and calcium. Most preferably R_2 has 12 to 14 carbon atoms, in a linear rather than branched chain.

Preferred anionic cleansing surfactants are selected from sodium lauryl sulphate and sodium lauryl ether sulphate(n)EO, (where n is from 1 to 3); more preferably sodium lauryl ether sulphate(n)EO, (where n is from 1 to 3); most preferably sodium lauryl ether sulphate(n)EO where $n=1$.

When included, preferably the level of alkyl ether sulphate is from 0.5 wt% to 25 wt% of the total composition, more preferably from 3 wt% to 18 wt%, most preferably from 6 wt% to 15 wt% of the total composition.

When included, the total amount of anionic cleansing surfactant generally ranges from 0.5 wt% to 45 wt%, more preferably from 1.5 wt% to 20 wt%.

Compositions of the invention may contain non-ionic surfactant. Most preferably non-ionic surfactants are present in the range 0 to 5 wt%.

Nonionic surfactants that can be included in compositions of the invention include condensation products of aliphatic ($C_8 - C_{18}$) primary or secondary linear or branched chain alcohols or phenols with alkylene oxides, usually ethylene oxide and generally having from 6 to 30 ethylene oxide groups. Alkyl ethoxylates are particularly preferred. Most preferred are alkyl ethoxylates having the formula $R-(OCH_2CH_2)_nOH$, where R is an alkyl chain of C12 to C15, and n is 5 to 9.

Other suitable nonionic surfactants include mono- or di-alkyl alkanolamides. Examples include coco mono- or di-ethanolamide and coco mono-isopropanolamide.

Further nonionic surfactants which can be included in compositions of the invention are the alkyl polyglycosides (APGs). Typically, APG is one which comprises an alkyl group connected (optionally via a bridging group) to a block of one or more glycosyl groups. Suitable alkyl polyglycosides for use in the invention are commercially available and include for example those materials identified as: Oramix NS10 ex Seppic; Plantaren 1200 and Plantaren 2000 ex Henkel.

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Other sugar-derived nonionic surfactants which can be included in compositions of the invention include the C_{10} - C_{18} N-alkyl (C_1 - C_6) polyhydroxy fatty acid amides, such as the C_{12} - C_{18} N-methyl glucamides, as described for example in WO 92/06154 and US 5,194,639, and the N-alkoxy polyhydroxy fatty acid amides, such as C_{10} - C_{18} N-(3-methoxypropyl) glucamide.

Amphoteric or zwitterionic surfactant can be included in an amount ranging from 0.5 wt% to about 8 wt%, more preferably from 1 wt% to 4 wt% of the total composition.

Examples of amphoteric or zwitterionic surfactants include alkyl amine oxides, alkyl betaines, alkyl amidopropyl betaines, alkyl sulphobetaines (sultaines), alkyl glycinate, alkyl carboxyglycinates, alkyl amphotacetates, alkyl amphopropionates, alkylamphoglycinates, alkyl amidopropyl hydroxysultaines, acyl taurates and acyl glutamates, wherein the alkyl and acyl groups have from 8 to 19 carbon atoms. Typical amphoteric and zwitterionic surfactants for use in shampoos of the invention include lauryl amine oxide, cocodimethyl sulphopropyl betaine, lauryl betaine, cocamidopropyl betaine and sodium cocoamphoacetate.

A particularly preferred amphoteric or zwitterionic surfactant is cocamidopropyl betaine.

Mixtures of any of the foregoing amphoteric or zwitterionic surfactants may also be suitable. Preferred mixtures are those of cocamidopropyl betaine with further amphoteric or zwitterionic surfactants as described above. A preferred further amphoteric or zwitterionic surfactant is sodium cocoamphoacetate.

The invention will now be illustrated with reference to the following non-limiting examples.

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Examples

1. Experimental Methods

5 1.1 Cell Culture

Primary human neonatal keratinocytes were purchased from Lonza, and stored in the vapour phase of liquid nitrogen until use. The cells were revived following the manufacturer's instructions and transferred into 20ml of fully supplemented KGM-Gold™ medium (Lonza) containing 70μM calcium in a T175cm² tissue culture flask (BD Falcon).

10 The cells were cultured following the manufacturer's instructions until approx 70% confluent. The cells were then sub-cultured into 12 well tissue culture plates (BD Falcon) in KGM-Gold Medium™ with 70μM calcium, but without hydrocortisone and GA-100 antibiotic /antimycotic supplements, and incubated for 24hrs before the treatments were applied.

15

1.2 Cell Treatment

Climbazole supplied by Symrise (Crinipan® AD) was dissolved into the solvent, dimethyl sulfoxide (DMSO) (Sigma Aldrich) to make stock solutions of 0.1 mM, 1mM and 10mM.

The stock solutions were diluted 1:1000 in KGM-Gold™ medium supplemented with
20 70μM calcium but containing no hydrocortisone or GA-100, to give final concentrations of climbazole of 0.1 μM, 1μM and 10μM. A solvent control of 0.001% DMSO was also prepared. Treatments were applied in triplicate, with 1.5ml of treatment per well. Cells were then incubated at 37°C, 5% CO₂ and incubated for either 24hr or 48hr, after which time the RNA was extracted. The cultured cells had reached approximately 80-90%
25 confluence after 48hr.

1.3 RNA Isolation & Analysis

RNA samples from the cells were prepared using an RNAeasy™ micro kit (Qiagen Ltd) following the manufacturer's instructions. The medium was removed from the cells, and
30 the cells were then washed briefly with phosphate buffered saline (DPBS) (Gibco Life Technologies Ltd) at room temperature. The cells were then lysed in buffer RLT before the sample was applied to a Qiashredder™ spin column (Qiagen Ltd). The lysate was used to extract RNA following the protocol provided, final elution was made into 14μl of RNase-free water and the samples stored at -80°C until further analysis. The quantity

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and quality of RNA extracted from the 24 samples was analysed using the Agilent 2100 Bioanalyzer™ (Agilent Technologies Ltd).

1.4 Microarray whole human genome gene expression analysis

5 Agilent SurePrint G3 Human Gene Expression 8x60K v2 Microarrays (Design ID 039494), which offer comprehensive coverage of ~50K human gene and transcript sequences, were used in conjunction with Agilent SureHyb hybridization and SureScan High-Resolution Technology in accordance with the established protocol relevant at the time of the study. Genome-wide gene expression analysis was carried out in accordance
10 with the Agilent One-Color Microarray-Based Gene Expression Analysis Protocol. Key consumables were purchased from Agilent Technologies UK Ltd unless otherwise noted.

For each sample, 300ng of RNA and the appropriate amount of Agilent One-Color RNA Spike-In RNA was labelled with reagents supplied in the Quick Amp Labeling Kit, one-
15 color according to manufacturer's instructions as follows: Primers were annealed to template by mixing 1.2 µl T7 Promoter Primer, RNA and spike-in control in a volume of 11.5 µl, denatured at 65°C for 10min followed by incubation at 4°C for 5min. First Strand Reaction Buffer (to 1X), DTT (to 10mM), dNTP (to 0.5mM), MMLV-RT (1 µl) and RNaseOut (0.5 µl) were then added to make up the cDNA synthesis reaction (20 µl),
20 which was incubated for 2h at 40 °C. After 15 min denaturation at 65°C, the reaction was made up to 60 µl with the addition of Cyanine 3-CTP (to 0.3mM), Transcription Buffer (to 1X), DTT (to 10mM), NTP (8 µl), PEG (to 4%), RNaseOUT (0.5 µl), Inorganic Phosphatase (0.6 µl), and T7 RNA Polymerase (0.8 µl) and incubated for 2h at 40 °C to synthesise the fluorescent labelled cRNA. The labelled cRNA was purified with the RNeasy Mini Kit
25 (Qiagen Ltd) according to manufacturer's protocol, and quantified according to Agilent's protocol.

The labelled cRNA was then hybridised to the Agilent SurePrint G3 Human Gene Expression 8x60K v2 Microarray slides using reagents from the Agilent Hi-RPM Gene
30 Expression Hybridization Kit according to manufacturer's protocol: 600ng of the labelled cRNA was used for hybridisation. Hybridisation was performed for 17 hr at 65 °C with 10 rpm rotation. Slides were then washed for 1 min at 22 °C in Wash Solution 1, followed by 1 min in Wash Solution 2 that was pre-warmed to 37 °C, 10s in acetonitrile (Fisher), and 30s in Agilent Stabilisation and Drying Solution. Slides were scanned with the Agilent

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Technologies Microarray Scanner G2505C US84700251 . Data was extracted from scanned microarray images using the Agilent Feature Extraction Software (v1 0.7.3.1).

2 Microarray Data Collection & Analysis

5 Data analysis was carried out using Qlucore Omics Explorer and GeneSpring GX12.

2.1 Qlucore Analysis

Initial analysis of the microarray datasets was performed using the Qlucore Omics Explorer software (Qlucore Ltd) to generate a principal component analysis (PCA) plot.

10 Samples and variables that visually appear close to each other in a PCA plot are usually also close in biological characteristics. Therefore, if clear differences are observed between the data sets in a PCA plot, it suggests the treatment has had a biological effect. The three dimensional PCA plot generated for the climbazole treated keratinocyte transcriptomic data is shown in Figure 1, with each datum point corresponding to a single
15 sample

The PCA plot included herein is a 2D representation of the 3D image generated by the Qlcore analysis. An explanation of the usefulness of this analysis technique is described in Kalocsai P., and Shams S., Journal of Laboratory Automation October 1999, vol. 4 no.
20 5, 58-61 . The PCA plot suggested that the microarray data generated were of high quality and there was clear evidence that the climbazole treatment had affected the transcription of genes in the keratinocytes. The data showed indicated that the higher the dose and longer the treatment time, the greater the degree of change.

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2.2 GeneSpring™ Analysis

- Further analysis of the microarray data set was performed using GeneSpring analysis software (Agilent Technologies Ltd). The software was used to identify the probe sets
- 5 that were significantly changed by the climbazole treatment, compared to the vehicle control. Moderated t-tests were carried out on gene entities flagged by the feature extraction software as detected in all triplicates of at least one of the pairs of conditions being tested
- 10 In order to get an overall view of gene expression changes, the data was filtered to select for gene entities flagged by the feature extraction software as detected in all triplicates of at least one of the eight treatment conditions present in the experiment, then subjected to 2-way ANOVA analysis (FDR = <0.05 Benjamini Hochberg MTC) to identify gene expression changes as a result of duration and/or dose of climbazole treatment. This
- 15 identified 1361 genes to be differentially expressed due to dose of climbazole.
- GO Analysis in GeneSpring results interpretation workflow was used to identify the gene ontology groups that were statistically over-represented in the genes identified to have altered expression as a result of dose of climbazole. Climbazole was found to have had a
- 20 large effect for the epidermal differentiation gene ontology group. Table 1 identifies the

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specific genes whose transcription was upregulated by treatment with climbazole, and the protein they encode.

Gene	Protein
IL-1A	Interleukin 1 alpha
LCE3A	Late cornified envelope protein 3A
SPRR2F	Small proline-rich protein 2F
CDSN	Corneodesmosin
LCE3D	Late cornified envelope protein 3D
SPRR2B	Small proline-rich protein 2B
CNFN	Cornifelin
LCE3B	Late cornified envelope protein 3B
EREG	Epiregulin
SPRR2G	Small proline-rich protein 2G
SPRR2A	Small proline-rich protein 2A
LCE2C	Late cornified envelope protein 2C
LCE3E	Late cornified envelope protein 3E
SPRR2D	Small proline-rich protein 2D

5 Table 1 Epidermal differentiation genes that are upregulated in primary keratinocytes treated with climbazole

These genes encode proteins that relate to epidermal differentiation, specifically proteins relating to skin barrier strengthening, and/or barrier repair, so upregulation of these genes

10 by treatment with climbazole indicates beneficial effects in skin barrier strengthening and/or skin barrier repair.

3 Evidence for the link between skin barrier benefits and Upregulation of the specified genes

3.1 Cornified Envelope

The cornified envelope is the protective barrier of stratified squamous epithelia, and is
5 synthesised at the late stages of keratinocyte differentiation. The cornified envelope is
insoluble and approximately 15nm thick. Assembly starts with the formation of a scaffold
consisting of involucrin and envoplakin, subsequently other reinforcing proteins are
added, such as cystatin, elafin, loricrin and small proline-rich proteins (SPRRs). These
proteins serve as an attachment platform for the addition of specific lipids. The formation
10 of cornified envelope is described in Candi E, Schmidt R & Melino G; The cornified
envelope: a model of cell death in the skin., Nat Rev Mol Cell Biol 2005 Apr; 6(4):328-40,
and depicted in figure 2 therein.

3.2 Late cornified envelope (LCE) proteins

15 LCE proteins are cornified envelope precursors that have protein cross-linking function.
The *LCE* gene cluster is part of the epidermal differentiation complex present on human
chromosome 1q21. With eighteen members, the *LCE* gene family is divided into six
groups, *LCE1* to *LCE6*, based on similarities of amino acid sequence, genomic
organisation, and patterns of expression. Investigators have used qPCR to demonstrate
20 that human *LCE1* and *LCE2* genes are primarily expressed in skin.

Work has been completed investigating changes in *LCE* gene expression in psoriatic skin
(see Bergboer J et al., Psoriasis risk genes of the late cornified envelope-3 group are
distinctly expressed compared with genes of other LCE groups, Am J Pathol 2011 Apr;
25 178(4):1470-7). Transcription of all members of the *LCE* gene groups 1, 2, 5 and 6 were
significantly downregulated in psoriatic skin, whereas all genes of the *LCE3* group, which
were barely present in normal skin, were significantly induced in psoriatic skin. qPCR
analysis was used to investigate expression of all *LCE* genes before and after tape
stripping (i.e. inducing barrier disruption/inflammation of normal skin). The *LCE* genes of
30 groups 1, 2, 5, and 6 were significantly downregulated after tape stripping, whereas the
LCE3 genes were significantly upregulated (Bergboer 2011). *In vitro* studies have
demonstrated that the pro-inflammatory cytokines associated with psoriasis e.g. TNF- α ,
IL-1 α and IL-6, significantly induce expression of *LCE3* genes. It is interpreted from
these studies that *LCE3* members function as barrier repair proteins whose expression is

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driven by pro-inflammatory cytokines, whereas other LCE proteins have a constitutive role in barrier function of normal skin.

It was shown in our microarray results that transcription of a number of the *LCE* genes was upregulated in climbazole treated primary keratinocytes. The majority of these changes were in *LCE* group 3 genes including *LCE3A*, *LCE3B*, *LCE3D*, *LCE3E* as well as the group 2 gene, *LCE2C*. Analysing these results in view of the literature, it is indicative of climbazole inducing changes in *LCE* gene expression suggestive of a barrier repair and/or strengthening effect.

3.3 Small proline-rich proteins

Human small proline-rich proteins (SPRRs) consist of a multi-gene family clustered within the epidermal differentiation complex on human chromosome 1q21. SPRRs have a specialised role and function as cross bridging agents reinforcing the cornified envelope.

Biochemical evidence has suggested that the characteristics of the cornified envelope related to toughness, strength and flexibility, exhibited by different stratified squamous epithelia, are dictated by the SPRR proteins (Cabral A et al., Structural organization and regulation of the small proline-rich family of cornified envelope precursors suggest a role in adaptive barrier function; J Biol Chem., 2001 Jun; 276(22):19231-7).

The results generated in our microarray analysis of climbazole treated keratinocytes showed a number of the class 2 *SPRR* gene transcripts were upregulated including *SPRR2A*, *SPRR2B*, *SPRR2D*, *SPRR2F* and *SPRR2G*. Analysing these results in view of the literature, it is indicative that climbazole is positively impacting on the strength and flexibility of the skin barrier.

3.4 Corneodesmosin

Corneodesmosin, and additional cornified envelope protein was also upregulated by climbazole treatment of keratinocytes.

Corneodesmosin (CDSN) is synthesised by granular keratinocytes and localises to specific cell junction structures. CDSN is incorporated into the desmoglea of the desmosomes, shortly before their transformation into corneodesmosomes during cornification. It is an important protein in maintaining the integrity of the epidermis (Jonca

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N et al., Corneodesmosomes and corneodesmosin: from the stratum corneum cohesion to pathophysiology of gendermatoses; Eur J Dermatol 201 1 May; 21 Suppl 2: 35-42).

5 Thus upregulation of this gene by treatment with climbazole is indicative that the skin barrier is being strengthened.

Conclusion of results

10 So the microarray results and analysis have shown that the use of climbazole has upregulated a number of genes involved in epidermal differentiation, specifically genes encoding cornified envelope proteins, small proline-rich proteins and corneodesmosin. As demonstrated in the literature, these proteins have a function for epidermal skin barrier benefits, especially skin barrier repair, and skin barrier strengthening.

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Claims

1. Cosmetic use of climbazole to provide an epidermal skin barrier benefit.
2. Cosmetic use according to claim 1, wherein the epidermal skin barrier benefit is
5 selected from: strengthening the skin barrier and/or repair of the skin barrier.
3. Cosmetic use according to claim 1 or claim 2, wherein the epidermal skin barrier benefit is a scalp skin barrier benefit.
- 10 4. Cosmetic use according to any preceding claim, wherein climbazole is used at a level of from 0.1 to 5 wt.%, preferably from 0.1 to 2.5 wt.% in a personal care composition.
- 15 5. Cosmetic use according to claim 4, wherein the personal care formulation is a hair and/or scalp treatment composition.
6. Cosmetic use according to claim 5, wherein the hair and/or scalp treatment composition is a shampoo, conditioner or a leave-on composition.
- 20 7. Cosmetic method for provision of an epidermal skin barrier benefit, comprising applying to the skin a composition comprising climbazole.
8. Cosmetic method according to claim 7, wherein the epidermal skin barrier benefit is selected from: strengthening the skin barrier and/or repair of the skin barrier.
25
9. Cosmetic method according to claim 7 or claim 8, wherein the epidermal skin barrier benefit is a scalp skin barrier benefit.
- 30 10. Cosmetic method according to any one of claims 7 to 9, wherein the composition comprises climbazole at a level of from 0.1 to 5 wt.%, preferably from 0.1 to 2.5 wt.%, and is a personal care composition, preferably a hair and/or scalp treatment composition.

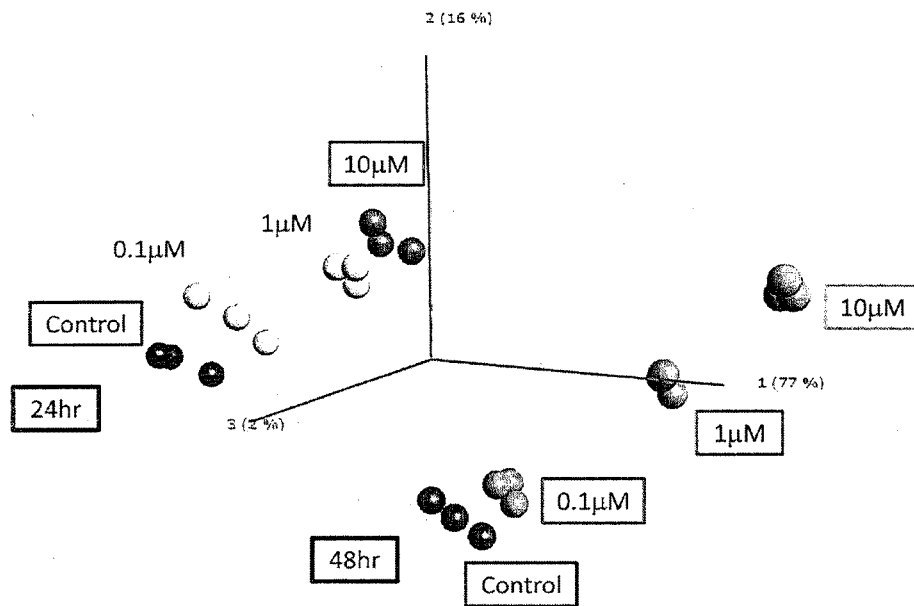
Drawings

Figure 1 PCA plot of transcriptomic data for climbazole treated keratinocyte samples

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2014/056082

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61Q5/02 A61Q5/12 A61K8/49
ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61Q A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

EPO-Internal , WPI Data, PAJ

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EP 0 803 248 A2 (UNI LEVER PLC [GB] ; UNI LEVER NV [NL]) 29 October 1997 (1997-10-29) page 3, line 1 - line 29 example 3 examples 7-9	1-10
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A	US 2008/268077 A1 (VI ELHABER GABRI ELE [DE]) 30 October 2008 (2008-10-30) the whole document	1-10



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Date of the actual completion of the international search

12 May 2014

Date of mailing of the international search report

22/05/2014

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INTERNATIONAL SEARCH REPORT

International application No

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International application No

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