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Deletion of the Late Cornified Envelope Genes *LCE3B* and *LCE3C* May Promote Chronic Hand Eczema With Allergic Contact Dermatitis

S Molin,[1] S Vollmer,[1] EH Weiss,[2] P Weisenseel,[1] T Ruzicka,[1] JC Prinz[1]

[1]Department of Dermatology and Allergology, Ludwig Maximilian University, Munich, Germany
[2]Department of Biology II, Ludwig Maximilian University, Munich, Germany

Abstract

Background: Genetically determined defects in epidermal skin barrier function may contribute to the development of irritant and/or allergic contact dermatitis in chronic hand eczema (CHE).
Objectives: To assess whether a deletion in the late cornified envelope genes *LCE3B* and *LCE3C* may constitute a genetic predisposition for the development of CHE or any of its subtypes.
Patients and Methods: A total of 153 German patients with clearly defined CHE subtypes and 268 healthy individuals were screened for the deletion *LCE3C_LCE3B-del* by allele-specific polymerase chain reaction.
Results: Classification of the patients by etiologic subtypes revealed an association between the *LCE3C_LCE3B-del* allele and CHE due to allergic contact dermatitis. In this subtype, 19/37 patients (51.4%) were homozygous deletion carriers, 11/37 (29.7%) were heterozygous carriers, and just 7/37 (18.9%) were wild-type individuals. Compared to the other CHE subgroups and the healthy control group (homozygous, 88/268 [32.83%]; heterozygous, 133/268 [49.63%]; and wild-type, 47/268 [17.54%]), the prevalence of *LCE3C_LCE3B-del* in these patients reached statistical significance (*P*=.03977), as did homozygous deletion carrier status (*P*=.01044 for other subtypes and *P*=.02695 for controls).
Conclusions: A deletion of *LCE* genes may promote the development of allergic contact dermatitis, which is a form of CHE involving delayed-type hypersensitivity.

Key words: Chronic hand eczema. Susceptibility. Late cornified envelope. Allergic contact dermatitis. Epidermal skin barrier function.

Resumen

Antecedentes: Los defectos genéticamente determinados en la función de barrera de la epidermis pueden contribuir a la aparición de dermatitis alérgica y/o irritante de contacto en el eccema crónico de manos (ECM).
Objetivos: Evaluar si la deleción de los genes *LCE3B* y *LCE3C* (*late cornified envelope*) puede constituir una predisposición genética al desarrollo de ECM o alguno de sus subtipos.
Pacientes y métodos: Un total de 153 pacientes alemanes con subtipos de ECM claramente definidos y 268 voluntarios sanos se sometieron a pruebas para la detección de la deleción *LCE3C_LCE3B-del* mediante la reacción en cadena de la polimerasa específica de alelo.
Resultados: La clasificación de los pacientes según el subtipo etiológico reveló una asociación entre el alelo *LCE3C_LCE3B-del* y el ECM debido a dermatitis alérgica de contacto. En este subtipo de ECM, 19/37 pacientes (51,4%) eran portadores homocigóticos de la deleción, 11/37 (29,7%) eran portadores heterocigóticos y solo 7/37 (18,9%) portaban el alelo natural. En comparación con los otros subgrupos de ECM y con el grupo de control de voluntarios sanos (homocigóticos, 88/268 [32,83%]; heterocigóticos, 133/268 [49,63%]; y alelo natural, 47/268 [17,54%]), la prevalencia de la deleción *LCE3C_LCE3B-del* en estos pacientes alcanzó significación estadística (*p*=0,03977), al igual que el estado de portador homocigótico de la deleción (*p*=0,01044 para otros subtipos y *p*=0,02695 para los controles).
Conclusiones: La deleción de genes LCE puede favorecer la aparición de dermatitis alérgica de contacto, que es una forma de ECM que cursa con hipersensibilidad de tipo retardado.

Palabras clave: Eccema crónico de manos. Susceptibilidad. *Late cornified envelope*. Dermatitis alérgica de contacto. Función de barrera de la epidermis.

Introduction

Chronic hand eczema (CHE) is commonly defined as a persistent, noninfectious skin inflammation with eczematous lesions restricted to the hands. The clinical spectrum extends from mild and transitory to severe and refractory eczema, with a high disease burden and a negative impact on many aspects of health-related quality of life [1-5]. CHE comprises a heterogeneous group of disease manifestations. Current classifications are based on etiologic distinctions (CHE due to contact sensitization, atopy, irritant exposure, or idiopathic forms) or morphologic distinctions (dyshidrotic or hyperkeratotic-rhagadiform skin lesions) [6]. All these forms are likely to involve a combination of endogenous predisposition and environmental triggers [7].

An essential etiological aspect of CHE is impaired epidermal skin barrier function, which may increase the penetration of allergens through the epidermis and reduce resistance to irritant damage. Skin barrier defects may result from genetically determined modifications in the structure of the cornified envelope [8-10]. The cornified envelope constitutes a physical barrier, which protects against water loss and the penetration of exogenous noxae [11]. Most of the proteins that make up this barrier are encoded by genes located in the epidermal differentiation complex (EDC) on chromosome 1q21; these genes are mandatory for keratinocyte differentiation and skin barrier maintenance. The EDC comprises 4 clusters of gene families: the late cornified envelope (*LCE*) family, the small proline rich protein (*SPRR*) family, the *S100* gene family, and the filaggrin (*FLG*) family [11-13]. A nonfunctional FLG mutation has been associated with CHE resulting from a combination of contact allergy and irritant skin damage [14]. *FLG* mutations may furthermore contribute to the development of irritant contact dermatitis (ICD) and allergic sensitization to nickel [15-16].

Alterations in *LCE* genes may also affect the functional state of the cornified envelope. Proteins encoded by these genes are integrated into the cornified envelope during epidermal differentiation or regeneration [12]. Although the physiological role of *LCE* genes is not yet clear, it has been proposed that they might be involved in epidermal skin barrier formation and repair [17-19]. A frequent deletion of the *LCE* genes, *LCE3B* and *LCE3C* (*LCE3C_LCE3B-del*), seems to affect the cornified envelope and is associated with various disorders including psoriasis [17-19].

We wondered whether a deletion of *LCE* genes might be associated with hand eczema in general, or, more specifically, with a certain CHE subtype related to impaired skin barrier function (eg, allergic contact dermatitis [ACD] or ICD). We therefore analyzed the frequency of *LCE3C_LCE3B-del* in a German cohort of 153 patients with CHE who had been classified into clearly defined clinical subtypes, and compared the results to those observed in 268 healthy controls.

The results of our analysis suggest that this deletion may promote the manifestation and persistence of ACD, which is a subtype of CHE characterized by delayed-type hypersensitivity.

Materials and Methods

Patients and Controls

We included patients with any etiologic or morphologic subtype of CHE. According to the German Guidelines on the Management of Hand Eczema [20], chronic disease was defined as the presence of skin lesions for more than 3 months or the occurrence of 2 or more relapses in 12 months. In total, 153 patients (78 men and 75 women with a mean [SD] age of 47.9 [15.8] years, range 17-80 years) were recruited between 2007 and 2009 at the Department of Dermatology and Allergology at the Ludwig Maximilian University in Munich, Germany. The patients were enrolled from polyclinic and outpatient departments as well as from inpatient wards. All patients gave their written informed consent for participation in this study, which was approved by the local ethics committee and conducted in accordance with the principles of the Declaration of Helsinki. Each patient underwent a detailed evaluation, which included an investigation of past medical history, seasonal or occupational variations, influence and degree of contact with water and other irritants, disease course, clinical manifestations, the presence of atopy, contact allergies, and a family or personal history of other skin diseases such as psoriasis. The survey was conducted using a detailed, purpose-developed, standardized questionnaire as previously described [14]. In total, 321 genomic DNA samples from students and employees at the Biological Faculty of the Ludwig Maximilian University were available and served as ethnically matched, unrelated healthy controls.

Genetic association studies usually require large cohorts of participants. A limiting factor in participant recruitment, though, is usually the number of cases available to study [21]. Although our patient group was limited in size, it offered a major advantage in that we had very detailed information on clinical course and disease symptoms. This is a premise for conducting reliable subtype analyses, particularly in etiologically and morphologically heterogeneous diseases such as CHE.

Classification of CHE

We evaluated the distribution and morphology of skin lesions, foot involvement, and clinical manifestations or signs of atopy according to the methods described by Rajka and Hanifin [22]. All the patients were screened for type I and type IV allergies by skin prick testing with common allergens (cat, house dust mite, and grass pollen), determination of serum immunoglobulin (Ig) E levels, and patch testing (using Finn chambers) with the current European standard series and additional series according to individual exposure [23]. All patients were assigned to a particular hand eczema subtype using a graphic diagnostic algorithm [24] consisting of ACD, ICD, combined ACD and ICD (AICD), each with or without atopy, as well as atopic and idiopathic CHE. This algorithm is based on common classifications for CHE and indicates the main criteria that should be used to distinguish between different subtypes of hand eczema.

A diagnosis of ACD was established by a positive patch test reaction for relevant contact allergens, a direct relationship with

allergen exposure or avoidance, and skin disease symptoms and eczema spreading beyond the hands. Excessive private or occupational hand contact with water or irritants served as a major criterion for the diagnosis of ICD. Patients with contact allergies and concomitant irritant damage were diagnosed as having AICD. Both ACD and ICD were seen in patients with and without atopy. Criteria for atopy included a presence or history of atopic disorders, atopic stigmata according to Hanifin and Rajka [22], elevated IgE levels, and/or positive prick tests for common allergens. Atopic CHE was diagnosed in patients with signs of atopy in the absence of irritant damage and contact allergy. Finally, patients without atopy, contact allergies, or irritant damage were diagnosed as having idiopathic CHE.

Three morphologic forms of CHE were distinguished: hyperkeratotic-rhagadiform lesions, dyshidrotic lesions, and a mixed pattern combining both types of lesions.

Common differential diagnoses such as psoriasis and fungal infection were excluded in all patients based on family and personal history and thorough clinical, microbiological, and, in unclear cases, histopathological examinations.

Genotyping

Genomic DNA was extracted from whole blood using standard methods (QIAamp, DNA Blood Maxi Kit, Qiagen, Hilden, Germany). Genotyping for the *LCE3C_LCE3B-del* variant was performed by the single-tube, allele-specific polymerase chain reaction method described by de Cid et al [17]. The procedure was blinded to clinical phenotype. The expected population allele frequencies according to the Hardy-Weinberg equilibrium were compared to our allele frequency findings to control for errors in genotyping. All 153 individuals in the patient group and the 268 individuals in the control group were successfully typed for *LCE3C_LCE3B-del*. This limited the study size to 1.75 healthy controls per patient, instead of the originally intended ratio of 2 to 1, which, for relatively common diseases, often provides the most efficient design [21].

Statistical Analysis

All features were analyzed bilaterally using the χ^2 test or the Fisher exact test (Java-using Münster Biometry Online-system, http://campus.uni-muenster.de/fileadmin/einrichtung/imib/lehre/skripte/biomathe/bio/bio.html), with statistical significance set at $P<.05$. Odds ratio and 95% confidence intervals were also calculated. Results with a significance level of $P\leq.10$ were considered to indicate tendency. Based on consultation with a statistician from the university for the same constellation in a previous study, *P* values are cited without correction for multiple testing due to the exploratory nature of our analysis. In other words, any significant results from our investigation have a primarily exploratory quality.

Individuals with missing data for the diagnostic criteria for the classification of CHE and the assignment of subtypes were excluded from the statistical analysis.

Results

Distribution of Clinical Phenotype and Diagnoses in Patients With CHE

In total, 153 patients with CHE participated in our study. ACD was diagnosed in 37 patients (24%), ICD in 32 patients (21%), AICD in 28 patients (18%), idiopathic CHE in 33 patients (22%), and atopic CHE in 23 patients (15%) (Table 1A).

Sixty-two (41%) of the patients had hyperkeratotic-rhagadiform lesions, 45 had dyshidrotic lesions (29%), and 46 (30%) had a mixed pattern (Table 1A). The clinical details of course of disease, manifestations, and environmental influences are shown in Tables 1A and 1B.

Ninety-two patients (60%) had persistent skin lesions and 57 (37%) had frequent relapses. Four individuals (3%)

Table 1A. Differentiation of Clinical Subtypes According to Etiology, Morphology and Allele Carrier Status in Patients With Chronic Hand Eczema (CHE)[a]

	Healthy Controls	CHE Patients	Allergic Contact Dermatitis	Irritant Contact Dermatitis	Combined Allergic and Irritant Contact dermatitis	Atopic Hand Eczema	Idiopathic CHE	Hyperkeratotic-Rhagadiform Lesions	Dyshidrotic Lesions	Mixed Pattern
Total No.	268	153	37/153	32/153	28/153	23/153	33/153	62/153	45/153	46/153
LCE3C_LCE3B -del homozygous	88/268	52/153	19/37	11/32	6/28	6/23	10/33	21/62	15/45	16/46
LCE3C_LCE3B-del heterozygous	133/268	67/153	11/37	16/32	17/28	11/23	12/33	26/62	20/45	21/46
Wild-type	47/268	34/153	7/37	5/32	5/28	6/23	11/33	15/62	10/45	9/46
At least 1 *LCE3C_LCE3B-del* allele	221/268	119/153	30/37	27/32	23/28	17/23	22/33	47/62	35/45	37/46

[a]Data shown as number of individuals.

Table 1B. Clinical Characteristics and Allele Carrier Status of Patients With Chronic Hand Eczema[a]

	No. of Patients	Persistent Course	Relapsing Course	Foot Involvement	Pruritus	Atopic Predisposition	Family History of Atopy	Relevant Contact Allergy	Palmar Localisation	Worsening With Psychosocial Stress	Excessive Contact With Water/ Irritants
Total. No. of Patients	153	92/153[b]	57/153[b]	64/153	122/153	41/153	36/153	65/153	115/153	60/153	128/153
LCE3C_LCE3B -del homozygous	52/153	31/92	18/57	23/64	42/122	23/41	9/36	25/65	41/115	20/60	44/128
LCE3C_LCE3B-del heterozygous	67/153	38/92	28/57	27/64	53/122	38/41	17/36	28/65	47/115	26/60	55/128
Wild-type	34/153	23/92	11/57	14/64	27/122	18/41	10/36	12/65	27/115	14/60	29/128
At least 1 *LCE3C_LCE3B*-del allele	119/153	69/92	46/57	50/64	95/122	61/41	26/36	53/65	88/115	46/60	99/128

[a]Data shown as number of patients.
[b]Four patients were not able to give information on the time course of their CHE.

were not able to differentiate between these states. Of the 153 patients, 64 (42%) also had eczematous skin lesions on the soles of their feet, with 28 (18%) having relevant contact sensitizations (eg, to potassium dichromate, rubber chemicals, acrylates, or vulcanization accelerators). Foot involvement was diagnostic for idiopathic hand eczema in the absence of atopy, contact allergies, or irritant damage of the feet. A majority of the patients with CHE (122/153, 80%) had pruritus. Sixty-five patients (42.5%) had relevant contact sensitizations; 37 (56.9%) of these had ACD and 28 (43.1%) had AICD. Sixty-three patients (41%) reported worsening of their condition with water contact, and 60 (39%) with psychosocial stress. In 115 patients (75%), the eczematous skin lesions mainly affected the palms, and 81 (53%) also had lesions in the interdigital spaces (Table 1B).

LCE3C_LCE3B Genotypes

The results of *LCE3C_LCE3B* genotyping were analyzed in consecutive steps. First, allele frequencies were compared between patients and controls (Tables 2A and 2B). Then, the different etiologic subtypes of CHE (ACD, ICD, AICD, and atopic and idiopathic hand eczema) were compared, as were the clinical phenotypes (Tables 1A and 1B), which were discriminated by clinical course or morphology.

Total Population of Chronic Hand Eczema Patients and Healthy Controls

In total, 119/153 CHE patients (77.78%) had at least 1 deleted allele (*LCE3C_LCE3B-del*); 43.79% (67/153) were heterozygous deletion carriers and 33.99% (52/153) were homozygous carriers (Tables 1A and 2A); the remaining 22.22% (34/153) were wild-type individuals. A comparable distribution of alleles was seen in the healthy control group (n=286), with 88 homozygous deletion carriers (32.83%), 133 heterozygous carriers (49.63%), and 47 wild-type individuals (17.54%) (Tables 1A and 2A).

Allele frequencies were found to be 0.5588 (55.88%) for the *LCE3C_LCE3B-del* allele and 0.4412 (44.12%) for the wild-type allele in the CHE group. In the healthy control group, they were 0.5765 (57.65%) for the *LCE3C_LCE3B-del* allele and 0.4235 (42.35%) for the wild-type allele. The differences

Table 2A. *LCE3C_LCE3B* Genotype Distribution in Patients With Chronic Hand Eczema (CHE) and Healthy Controls[a]

	LCE3C_LCE3B	
	Patients	Controls
aa	52 (33.99)	88 (32.83)
Aa	67 (43.79)	133 (49.63)
AA	34 (22.22)	47 (17.54)
Total	153	268

Abbreviations: aa, homozygous for deleted *LCE3C_LCE3B* allele; Aa, heterozygous for deleted *LCE3C_LCE3B* allele; AA, homozygous for wild-type *LCE3C_LCE3B* allele.
[a]Data shown as number (%) of patients.

Table 2B. Allele Frequencies in Patients With Chronic Hand Eczema (CHE) and Healthy Controls[a]

LCE3C_LCE3B	Patients	Controls
Wild-type	135 (0.4412; 44.12)	227 (0.4235; 42.35)
Deleted allele	171 (0.5588; 55.88)	309 (0.5765; 57.65)

[a]Data shown as number of alleles, allele frequencies and percentages.

were not significant (Table 2B). Neither the difference between the numbers of individuals carrying at least 1 deleted allele nor the relationship between deleted and wild-type alleles reached statistical significance for *LCE3C_LCE3B-del* (CHE patients with at least 1 deleted allele: *P*=.24082; OR, 0.74434; 95% CI, 0.44126-1.25707; number of deleted alleles in CHE patients: *P*=.61840, OR, 0.93053, 95% CI, 0.69393-1.24790).

Etiologic Hand Eczema Subtypes

In the next step we analyzed the occurrence of *LCE3C_LCE3B-del* alleles in each etiologic subtype of chronic hand eczema (Tables 1A and 1B).

In patients with ACD, 19/37 (51.4%) were homozygous deletion carriers, 11/37 (29.7%) were heterozygous carriers, and 7/37 (18.9%) were wild-type individuals. Compared to the non-ACD CHE patients, the prevalence of *LCE3C_LCE3B-del* alleles did reach statistical significance (*P*=.03977; OR, 1.76721; 95% CI, 1.0233-3.0518). Statistical analysis also confirmed the observation that in the ACD group, the number of homozygous deletion carriers (19/37; 51.4%) was significantly higher than in the non-ACD groups (33/116 [28.4%]; *P*=.01044; OR, 2.65488; 95% CI, 1.16167-6.09075) and in the healthy control group (88/268 [32.8%]; *P*=.02695; OR, 2.15909; 95% CI, 1.02361-4.55981) (Table 3).

Eleven out of 32 patients with ICD were homozygous deletion carriers (34.4%), 16 (50%) were heterozygous carriers, and 5 (15.6%) were wild-type individuals. In total, thus, 27/32 (84.4%) of these patients were deletion carriers. In the group of 28 patients with AICD, 6 (21.4%) were homozygous for *LCE3C_LCE3B-del*, 17 (60.7%) were heterozygous, and 5 (17.9%) were wild-type individuals: In total, thus, there were 23 deletion carriers (82.1%). In the atopic CHE group, 6/23 individuals (26.1%) were homozygous deletion carriers, 11 (47.8%) were heterozygous carriers, and 6 (26.1%) were wild-type individuals (total of 17 deletions carriers [73.9%]). Finally, in the group of patients with idiopathic hand eczema (n=33), 22 (66.7%) were deletion carriers. Specifically, there were 10 homozygous carriers (30.3%), 12 heterozygous carriers (36.4%), and 11 wild-type individuals (33.3%).

Neither the number of deletion carriers nor the prevalence of *LCE3C_LCE3B-del* was statistically significant for ICD, AICD, or atopic or idiopathic CHE compared to each other.

Thus, statistical significance for the prevalence of the *LCE3C_LCE3B-del* allele was reached for individuals with CHE due to contact sensitization (ACD) when compared to the other CHE subgroups.

Morphologic Subtypes and Clinical Features of CHE

None of the morphologic phenotypes was clearly associated with the prevalence of the *LCE3C_LCE3B-del* allele (Table 1B). Twenty-one (33.9%) out of 62 individuals with hyperkeratotic-rhagadiform skin lesions were homozygous deletion carriers, 26 (41.9%) were heterozygous carriers, and 15 (24.2%) were wild-type individuals. Thus, 47 (75.8%) of the 62 patients with hyperkeratotic-rhagadiform hand eczema were deletion carriers. In the dyshidrotic hand eczema group (n=45), 15 patients (33.3%) were homozygous, 20 (44.4%) were heterozygous, and 10 (22.2%) were wild-type individuals

Table 3. Homozygous *LCE3C_LCE3B* Deletion Carriers in Etiologic Chronic Hand Eczema (CHE) Subtype Groups and Healthy Control Group[a]

	Healthy Controls	Allergic Contact Dermatitis	Irritant Contact Dermatitis	Cmbined Allergic and Irritant Contact Dermatitis	Atopic Hand Eczema	Idiopathic CHE
LCE3C_LCE3B-del homozygous	88/268 (32.8)	19/37 (51.4)	11/32 (34.4)	6/28 (21.4)	6/23 (26.1)	10/33 (30.3)

[a]Data shown as number (%) of patients.

Table 4A. Distribution of Etiologic and Morphologic Subtypes in the 119 CHE Deletion Carriers

	CHE Deletion Carriers	Allergic Contact Dermatitis	Irritant Contact Dermatitis	Combined Allergic and Irritant Contact Dermatitis	Atopic Hand Eczema	Idiopathic CHE	Hyperkeratotic-Rhagadiform Lesions	Dyshidrotic Lesions	Mixed Pattern
No. of patients with at least 1 *LCE3C_LCE3B-del* allele	119	30/119	27/119	23/119	17/119	22/119	47/119	35/119	37/119

Abbreviation: CHE, chronic hand eczema.

Table 4B. Distribution of Clinical Characteristics in the 119 CHE Deletion Carriers

	CHE Deletion	Persistent Course	Relapsing Course	Foot Involvement	Pruritus	Atopic Predisposition	Family History of Atopy	Relevant Contact Allergy	Palmar Localization	Worsening With Psychosocial Stress	Increased Contact With Water/ Irritants
No. of patients with at least 1 *LCE3C_LCE3B-del* allele	119	69/119[a]	46/119*	50/119	95/119	31/119	26/119	53/119	88/119	46/119	99/119

Abbreviation: CHE, chronic hand eczema.
[a]Four patients were not able to give information on the time course of their CHE.

(total number of deletion carriers, 35 [77.8%]). Patients with a mixed pattern of skin lesions were found to be homozygous for *LCE3C_LCE3B-del* in 16/46 cases (34.8%), heterozygous in 21/46 cases (45.6%), and wild-type in 9/46 (19.6%) cases, This corresponds to a total of 37/46 deletion carriers (80.4%).

No statistically significant associations were observed between the different clinical parameters investigated in the CHE group and the prevalence of *LCE3C_LCE3B-del* (Table 1B). The 128 patients who had reported excessive water contact or irritant exposure comprised 44 homozygous deletion carriers (34.4%), 55 heterozygous carriers (43%), and 29 wild-type individuals (22.6%). In total, thus, there were 99 deletion carriers (77.4%) in this group. Twenty-three (29.1%) of the 79 patients with atopic predisposition were homozygous deletions carriers, 38 (48.1%) were heterozygous carriers, 18 (22.8%) were wild-type individuals, and 61 (77.2%) were carriers of at least 1 deleted allele. Foot involvement was observed in 64 patients, 23 (35.9%) of whom were homozygous carriers, 27 (42.2%) of whom were heterozygous carriers, and 14 (21.9%) of whom were wild-type individuals (total number of carriers of at least 1 deleted allele, 50 [78.1%]).

Mutation Carriers in All Study Populations

The 119 CHE patients carrying at least 1 deleted *LCE3C_LCE3B* allele (52/119 [43.7%] homozygous, 67/119 [56.3%] heterozygous) were analyzed in more detail (Tables 4A and 4B), but classification according to clinical subtypes and morphology showed no additional statistically significant associations (data not shown).

Discussion

CHE features etiologically different sub-diseases that can be distinguished by clinical manifestation, allergic status of the individual, and influence of environmental triggers. These differences must be considered when analyzing the role of potential candidate genes for CHE. In our patients, CHE was classified according to clinical features, causal factors, and morphology of skin lesions using a diagnostic algorithm [24]. These examinations also ruled out the possibility that the skin changes might be related to psoriasis. Because psoriasis, which has been related to *LCE3C_LCE3B* deletions in large cohort studies [17-19] might have confounded our analysis, patients in whom this disease could not be firmly excluded were excluded from the study.

While a similar proportion of *LCE3C_LCE3B* deletion carriers was found in the study group and the control group, significant differences were found for 1 particular CHE subtype. Specifically, in the group of patients with CHE due to allergic contact dermatitis, there was a statistically significant increased prevalence of the *LCE3C_LCE3B-del* allele (*P*=.03977; OR 1.76721; 95% CI, 1.0233-3.0518) and an increased frequency of homozygous *LCE3C_LCE3B-del* carriers (51.4%) compared to the other CHE subtypes and healthy controls. This is an interesting finding, because in addition to heterozygous *LCE3C_LCE3B* deletions, a complete loss of *LCE3C_LCE3B* might determine more severe epidermal skin barrier disruption. The pathogenesis of ACD requires allergens to penetrate the epidermis. Accordingly, a defect in the epidermal barrier due to a complete loss of the *LCE3C_LCE3B* gene might contribute to the sensitization process in predisposed individuals by enhancing the penetration of environmental molecules into the skin. The complete loss of *LCE3C_LCE3B-del* would therefore be consistent with a greater risk for sensitization.

An increased prevalence of the *LCE3C_LCE3B-del* allele has also been observed in psoriasis patients [17], yet this prevalence was lower than that found in our study for CHE patients with ACD. Furthermore, the proportion of homozygous *LCE3C_LCE3B-del* allele carriers found in the psoriasis group was slightly lower than the proportion of heterozygous deletion carriers (44.8% vs 45.6%) [17]. In addition, a large population-based German study had previously excluded an association between contact sensitization prevalence and psoriasis [25]. Therefore the association between homozygous carrier status for the deleted *LCE3C_LCE3B* allele and ACD in CHE might be the result of increased relevance. The main risk alleles in psoriasis are related to innate and adaptive immune mechanisms. Due to an increased penetration of substances such as bacterial toxins, an epidermal barrier defect due to an *LCE3C_LCE3B* deletion might act as a nonspecific proinflammatory stimulus and favor the specific causes of psoriasis. This, however, would not contradict a pathophysiological relevance of a *LCE3C_LCE3B-del*-related skin barrier defect in allergic CHE, which is dependent on the penetration of allergens into the skin.

The *LCE3C_LCE3B* deletion is the second genetic predisposition found for a particular subtype of hand eczema. The other one is related to filaggrin expression. Heterozygous nonfunctional *FLG* mutations seem to confer weaker skin barrier resistance and promote the development of contact sensitization only in the presence of environmental barrier disruption such as excessive water contact, leading to CHE with AICD [14]. In contrast to the case of *FLG*, our data suggest that the effect of the *LCE3C_LCE3B* deletion on allergen penetration is independent of external irritant damage. A deletion of LCE genes may thus be an important cofactor for the development and persistence of ACD in CHE.

Although atopy is assumed to be a major individual risk factor for hand eczema [6-7], in our patient group, no association was found between atopic predisposition or atopic hand eczema with the *LCE3C_LCE3B* deletion. This observation supports the current perception that a genetic predisposition for hand eczema is mostly independent of coexistent atopic eczema, and is in agreement with recent data, which have excluded an association between *LCE3C_LCE3B* deletion and atopic dermatitis [12,26]. Interestingly, no association was observed between *LCE3C_LCE3B* deletion status and morphologic phenotypes of CHE. This may indicate that the etiology and clinical manifestations of CHE (morphology) depend on different traits.

One limitation of our findings is that our study and statistical results are of an exploratory nature. Further validation of our results by future studies of the association between CHE and ACD and the *LCE3C_LCE3B* deletion is warranted. The number of patients investigated was limited, as was the control group. Additional studies with larger study populations are required to clearly delineate the tendency for the statistically significant association shown by our data.

In conclusion, in addition to what is already known about FLG mutations, the results of this study provide further clues regarding genetic predisposition in terms of the etiology of the heterogeneous disease entity CHE. They suggest that individual variations in skin barrier function may be key to the development of particular forms of this disease. The analysis of additional gene families in the EDC might identify additional genetic variations which promote the development and persistence of CHE.

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S. Molin

Department of Dermatology and Allergology
Ludwig Maximilian University
Frauenlobstrasse 9 -11,
D- 80337 Munich
Germany
E-mail: sonja.molin@med.uni-muenchen.de

