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Dual role of CARD14 in skin inflammatory diseases

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Pursuing the Unknown

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Introduction

- Atopic dermatitis (AD) is a highly prevalent chronic inflammatory skin disease which is known to be, at least in part, genetically determined.
- Mutations in *CARD14* have been shown to result in various forms of psoriasis and related disorders.
- In this study we aimed to identify rare DNA variants conferring a significant risk for AD through genetic and functional studies in a cohort of patients affected with severe atopic dermatitis.

Clinical features and mutation analysis

We studied three families with index patients suffering from severe atopic dermatitis. Affected individuals displayed (A) erythroderma with fine scales involving more than 90% of their body surface area, and (B) prominent fissuring and accentuation of the skin folds; (C) Histopathological examination revealed psoriasiform epidermal hyperplasia and spongiosis. Using whole-exome and direct sequencing analysis, we identified 2 heterozygous missense mutations in *CARD14*. Individual II-1 from family 1 and individual II-1 from family 2 were found to carry the same heterozygous missense mutation in *CARD14* (c.1778T>C, p.I593T), whereas individuals II-1 from family 3 was found to carry a different heterozygous missense mutation (c.2209A>C, p.N737H). Both mutations are foreseen to be pathogenic and are in the structural PDZ-SH3-GUK module, also known as membrane-associated GUK(MAGUK) domain. This domain is essential for proper *CARD14* function, which suggests that the mutation may affect *CARD14* capacity to regulate NF- κ B signaling.

CARD14 mutations effect on NF- κ B signaling

We used NF- κ B luciferase reporter system to evaluate the effect of the mutations in *CARD14* on NF- κ B activity. HEK293 cells were co-transfected with NF- κ B luciferase reporter and with *CARD14*-wildtype, *CARD14*-mutants or a combination of these expression vector. Both mutations significantly attenuated the ability of *CARD14* to activate NF- κ B. Co-transfection of mutant and wild-type *CARD14* expression vectors led to impaired luciferase activity, indicating that the loss-of-function variants exert a dominant-negative effect. In accordance, we found a significant decrease in the expression of both *CARD14* and the activated p65 subunit of NF- κ B, in patients compared to healthy controls as detected in immunohistochemistry.

CARD14 mutations effect on antimicrobial peptides

CARD14 is a major regulator of NF- κ B signaling, which regulates innate immunity. We therefore examined the expression of pivotal mediators of innate immunity in human keratinocytes (KC) with knock-down *CARD14* or KC overexpressing mutant-*CARD14*. Both KC with reduced *CARD14* expression or mutant *CARD14*-overexpression released significantly less hBD1, hBD2 and hCCL20. Accordingly, the expression of these proteins was markedly reduced in the epidermis of our patient carrying a loss-of-function *CARD14* mutation as compared with healthy controls and psoriasis patients.

Conclusions

- Psoriasis and AD can be caused by opposite mechanisms associated with abnormal function of *CARD14* and consequent dysregulation of NF- κ B activity, pointing to a critical role of *CARD14* in the regulation of cutaneous inflammation.
- Decreased activity of *CARD14* signaling leads to down-regulation of anti-microbial peptide secretion, which in turn may underlie the susceptibility of these and other AD patients to infections.