Exploring the role of atopy in non-atopic diseases

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General introduction

ATOPY

Knowing the atopy status of patients is considered valuable information in dermatologic daily clinical practice. As a rule, in all patients presenting with a pruritic or an inflammatory skin condition, history of atopy is taken and documented. The importance of this information has been recognized and taught to residents for decades. The dermatologists know by experience that an atopic predisposition may be a clue to the diagnosis or a direct explanation for the patient´s complaints. In epidemiologic terms, the dermatologist is assessing for the risk factor 'atopy' and including or ruling out its comorbidities. Despite its daily use, the exact meaning of the term "atopy" has not been precisely delineated and the research into its comorbidities has not been approached in a systematic way.

The word "atopy" comes from the Greek word *ἀτοπία* which literally means "placelessness" but was intended to roughly mean "extraordinary" or "unusual". The term was coined by Coca and Cooke in 1923.[1] At that time it designated all immunologic phenomena which do not contribute to the body defenses against pathogens. Since then atopic disorders were thought to represent hypersensitivity reactions i.e. reactions of the immune system against harmless antigens. After the discovery of the Immunoglobulin E (IgE) in 1966 the term atopy was reserved for disorders in which IgE played a direct pathophysiological role.[2] Patients with atopic disorders, asthma, hay fever and atopic dermatitis (AD) were thought to be predisposed to producing IgE against various antigens such as house dust mite proteins, cat dandruff and pollen. As this is the case with many atopic individuals, some experts argue that the term "atopic" should be reserved for the ones exhibiting increased levels of serum IgE. In recognition of the heterogeneity of asthma and atopic dermatitis, new nomenclature had been developed in order to differentiate between "intrinsic", without, and "extrinsic", with increased IgE levels variants of these diseases.[3] Currently "atopy" has come to mean: a heritable constitution which predisposes individuals to the development of asthma, food allergies, hay fever and AD. These four distinct, comorbid conditions are subject to paradigm shifting and continually evolving definitions. Currently all four diagnoses are made by meeting validated, clinical disease criteria. However, no overall criteria for "atopy "exist. Besides the strongly associated atopic disorders, atopy is also considered to be a risk factor for various non-atopic conditions. The main emphasis of this thesis is on the association of AD with non-atopic conditions.

ATOPIC DERMATITIS

Epidemiology

AD is also known as eczema, atopic eczema, Besnier's prurigo, infantile eczema, flexural eczema, neurodermatitis and "constitutioneel eczeem" in Dutch. AD is usually the first atopic condition to be seen in an atopic individual and brings the atopic individuals to the attention of dermatologists. AD is also the most common inflammatory skin disease affecting up to 30% of infants and 10% of adults with its incidence tripling over the past three decades.[4-6] AD is typically a disease of the infancy as more than 50% of the patients develop the disease before the age of 6 months. Up to 70% of all infant cases of AD remit by the age of 7.[7] Risk factors for the development of AD are a family history of AD and other atopic disorders, exposure to bacterial endotoxins during the first year of life, exposure to antibiotics and decreased diversity of the gut microbiota. Protective factors are day care attendance, exposure to farm animals, consumption of unpasteurized milk and exposure to dogs. [8]

Pathophysiology

Although the AD pathophysiology is multifactorial and very complex its pathogenetic pathways can be roughly categorized in to epidermal barrier defects and aberrations of the adaptive immune system.[9] In short, the disturbances in the skin barrier lead to transepidermal water loss and xerosis but also allow penetration of antigens and toxins which then activate other parts of the innate and the adaptive immune system. As a consequence, inflammation and even further degradation of the skin barrier follow, presenting with classic signs of dermatitis in the skin.

One of the crucial barrier genes implied in the development of AD is the filaggrin gene (FLG).[10,11] The most common mutations in whites are R501X and 2284del4 and are found in 50% of severe and 30% of all AD cases. Some rare mutated variants of this gene increase the odds of AD by 151 fold.[12] The FLG codes for a protein called profilaggrin which after proteolysis becomes filaggrin and provides molecular scaffolding for the epidermal keratinocytes and contributes to the compaction of the stratum corneum and cohesion between its constituents.[13] Filaggrin cleavage by caspase-14 is required for its proper functioning.[14] A major derivative of this cleavage process is urocanic acid.[15,16] Urocanic acid provides protection against dehydration and low-level protection against UVB-induced DNA-damage.[17,18]Nevertheless, as only 30% of European patients with atopic dermatitis are a carrier of FLG mutations, deficiencies in other epidermal barrier constituents probably play an additional role.[19-21] There is evidence that the dysfunctions in other components of the epidermal barrier such as the cornified envelope, intercellular lipid lamellae, corneodesmosomes and corneocyte desquamation also play a role.[22,23]

Nevertheless, it was the discovery of the specific role of filaggrin in 2006 that shifted the pathophysiological paradigm to epidermal barrier dysfunction as the initiating step.[24] This sparked a multitude of novel research on filaggrin producing increasing numbers of papers. Mutations in the FLG gene are not only a risk factor for severe and persistent AD but also increase the risk of asthma by 3 times in the presence of eczema. [25,26] After the disruption of the epidermal barrier which is considered a failure of the first line of the innate defenses, other innate and adaptive parts of the immune system drive the pathophysiology. The inflammation involved is orchestrated by the Th2 lymphocytes through thymic stromal lymphopoetin (TSLP) and the cytokines IL-4, IL-5, IL-13 and IL-22.[27,28] In the chronic phase the Th1 and the Th17 axes are also involved.[29] There is a buildup of evidence of the involvement of the recently discovered lineage of group 2 innate lymphoid cells (ILC2s). Just like Th-2 lymphocytes the ILC2s produce large amounts of Th-2 cytokines, stimulate IgE production and show increased counts in AD, asthma and hay fever.[30] There is also an important role for microbes. Staphylococci, found in the lesional skin of up to 90% of the AD patients are also thought to add to this inflammatory mixture by further degrading the barrier by proteolytic enzymes and through the elicitation of immune responses by their enterotoxin.[31] With the help of the staphylococcal protease, the staphylococci penetrate the epidermis leaving a trail of inflammation characterized by increased secretion of IL-4, IL-13, IL-22 and TSLP while inhibiting the production of Th-1 cytokines and chemokines normally responsible for the coordination of clearance of staphylococcal infections.[32,33] The presence of staphylococci at least perpetuates the inflammation but may also be the major initial cause of AD.[34,35]

From AD to a system disease

A serologic marker of atopy in AD is the IgE anti-body. Eight to 80% of patients with AD exhibit increased levels of IgE depending on whether the study was community or hospital based.[36] Raised IgE correlates positively with AD severity and a worse long term prognosis. Though many AD patients may exhibit increased serum IgE levels, the IgE-repertoire in AD patients is much more restricted than in patients with asthma or hay fever which is suggestive of sensitization by a superantigen.[37] Furthermore, autoreactivity to proteins homologue to environmental antigens may also play a role.[38] Several other potential serum markers of AD exist such as thymus and activation regulated chemokine (TARC), T-cell attracting chemokine (CTACK), E-selectin, macrophage-derived chemokine(MDC), LDH and IL-18.[39,40] Recently α-defensin has been found to be increased during flare-ups of AD.[41] Although all four atopic diseases take stage at the interface between the body and the outside world the barrier dysfunction in the skin seems to precede and influence the onset of asthma, hay fever and food allergy (table 1).

One of the peculiarities of the atopic constitution is that patients may develop any and all of the four atopic disorders while their onset usually takes place in a fixed order. This specific and strong serial clustering of comorbidities is called the atopic march.[42] In practice, the atopic march means that an infant with AD could develop food allergies concomitantly and may go on to become asthmatic during childhood and later on during adolescence get affected by hay fever (figure 1). Subsequently, the atopic patient could present to various specialties with complaints related to any of these conditions. In addition, the rising incidence of atopy makes it a significant health problem. Taken the foregoing into consideration, atopic disorders make for a very interesting and challenging field for the study of comorbidities, be it from an epidemiologic or an immunologic point of view.

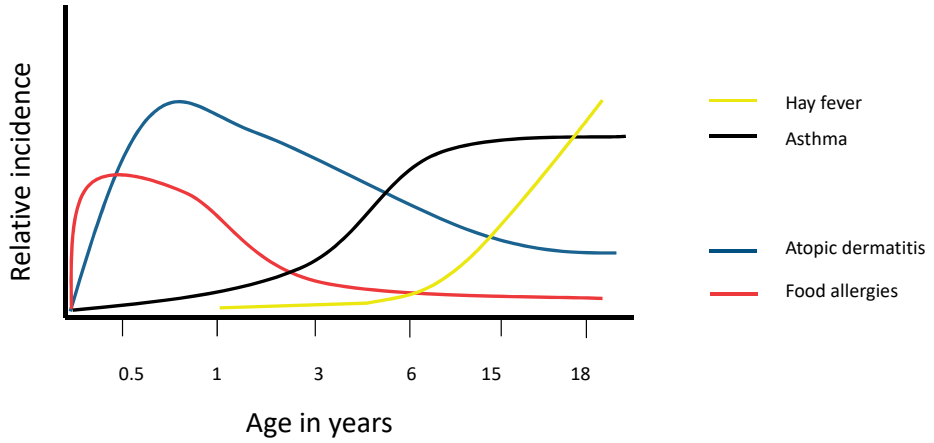

Figure 1. The course of the atopic march.

Clinical presentation

Patients with AD classically present with itch (pruritus), skin lesions and dry skin. The dermatologic findings typically vary with age. In infants AD usually presents with erythema, weeping and serous crusting on the face (figure 2).

On the trunk there may be red edematous papules and vesicles. The flexures of the neck and scalp may be involved in infants. During childhood lichenification sets in and lesions typical of AD can be found in antecubital and popliteal flexures. In adults more hand and periorbital dermatitis can be found. Persistence of AD in adulthood is more likely in patients with early onset, hay fever and hand dermatitis during childhood.[43] AD is therefore a polymorphous cutaneous disease which presents with multiple efflorescences at the same time (figure 3)

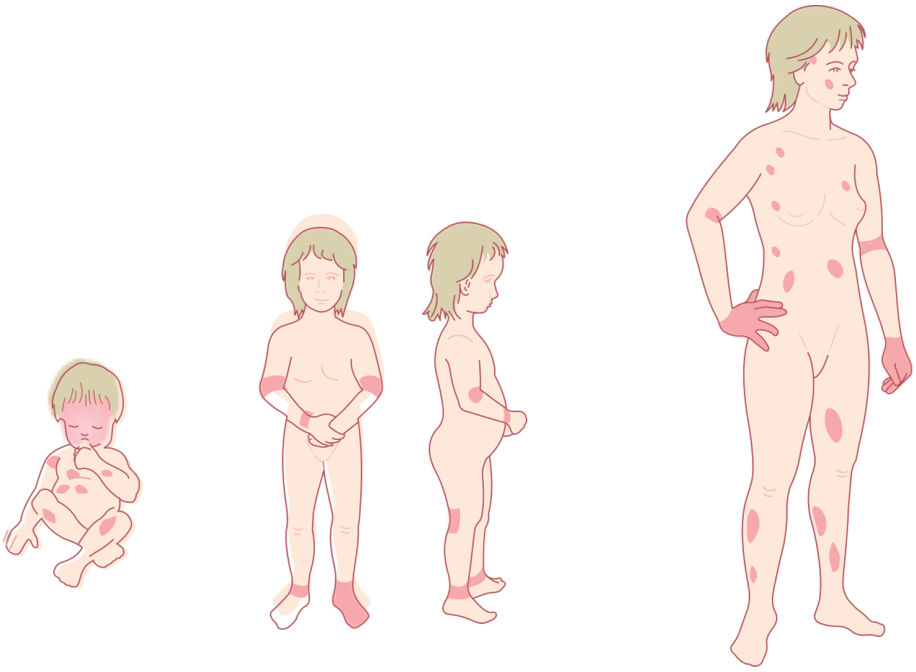

Figure 2. Evolution of atopic dermatitis from infant and child to adolescent

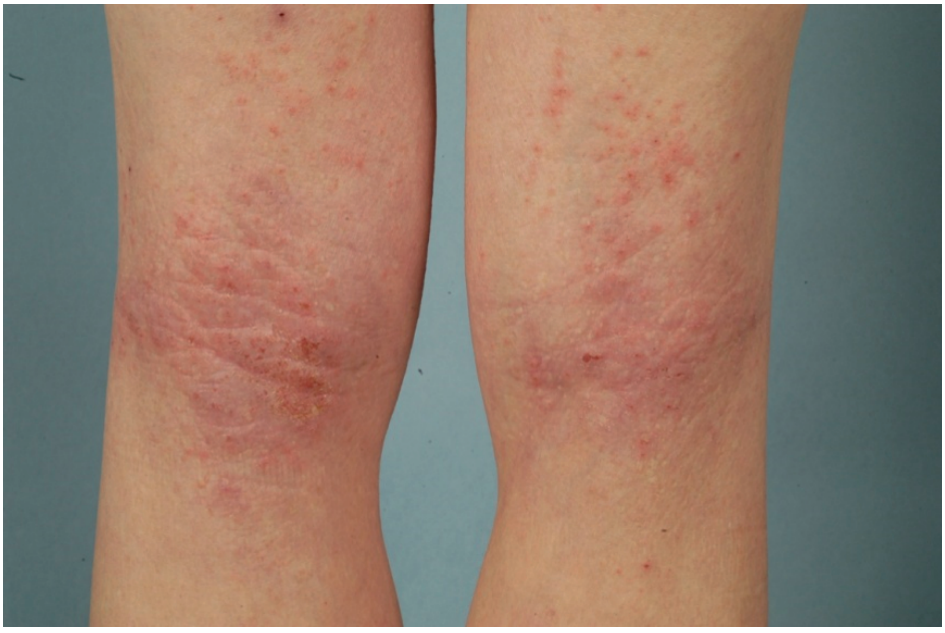

Figure 3. Atopic dermatitis of the popliteal flexures. There a papules, vesicles, lichenification, hemorrhagic and serous crusting, excoriations, vaguely demarcated erythema and scaling.

Table 1. Illustration of the anatomic localization of atopy according to the specific disease

Atopic disease	Involved body interface
Atopic dermatitis	Skin
Asthma	Respiratory lining
Hay fever	Conjunctiva and nasal mucosa
Food allergy	Oral mucosa, esophageal mucosa, gut lining

The diagnosis is made by symptoms and clinical findings only. The diagnosis of AD is usually made according to the diagnostic guidelines for atopic dermatitis by the U.K. working party which is the most validated diagnostic tool in the clinical and investigational settings.[44] These criteria comprise one major criterion plus three or more minor criteria. 'An itchy skin condition in past 12 months' is the must have criterion and the participant needs to fulfil at least three minor criteria which are: 'history of involvement of the skin creases', 'a personal history of asthma or hay fever', 'a history of generally dry skin in the last year', 'visible flexural eczema' and 'onset under 2 years of age' (table 2)

Table 2. UK working party's AD criteria

Major	Itchy skin condition past 12 months
Minor	Personal or 1st degree relative history of asthma or hay fever
	History of involvement of skin creases
	History of generally dry skin
	Visible flexural dermatitis
	Onset below age of 2

The major criterion is obligatory and the patient must fulfil at least 3 minor criteria.

Treatment

The treatment of AD consist of two pillars: restoration of the epidermal barrier and halting of the inflammatory reaction.[45] The first is achieved by avoidance of barrier damaging agents such as excessive use of soaps and water and applying generous quantities of emollients. Irritants such as wool, sweating and chlorine should also be avoided.[46] The second part is achieved with topical corticosteroids in most cases.[47] Application of these medications quickly reduces the pruritus and the inflammation and helps to restore and maintain the skin barrier at a very low risks of adverse events. In the past two decades topical calcineurin inhibitors have found their way in to the dermatologic arsenal for AD and prove to be a good alternative in some AD cases.[48] Another emerging topical treatment with good effect are inhibitors of phosphodiesterase-4.[49] However in the topical class the corticosteroids remain unchallenged in terms of efficacy, adverse events profile and cost effectiveness. Unfortunately in moderate and severe cases of AD, topical treatment only may not be sufficient. Treatment with UVB is then usually successful but

cannot be continued indefinitely.[50] Short or long courses of oral corticosteroids can be considered with or without the addition of azathioprine.[51] Other immunosuppressants which have been shown to be effective are ciclosporin A, mycophenolate mofetil and methotrexate.[52-54] Dupilumab, a monoclonal antibody that blocks IL-4 and IL-13 has shown very promising results but is as of now not registered in the Netherlands.[55,56] More biologic treatments are expected to be developed in the near future. Besides pharmacologic treatment, counseling, patient education and even psychologic care are often necessary.[57]

ATOPIC DIATHESIS AND COMORBIDITIES

Background

Comorbidity is the co-occurrence of two or multiple diseases in one patient. It is associated with worse clinical outcomes, increased health costs and more complex clinical management. [58] The construct of comorbidity is used differently in the fields of clinical care and public health. In the clinical sense a comorbid disease may or may not share the same pathogenesis or may even be a "complication" of the index disease. The comorbidities may or may not associate more frequently than is expected on the basis of chance. When this is the case however, there is an association or sharing of risk factors. Research into comorbidities provides valuable knowledge of the etiological models of disease as well as potential prevention approaches. The emphasis of this thesis is on AD. Dermatologic research on this topic however, cannot be conducted without accounting for the highly associated asthma, hay fever and food allergies which are all part of the atopic diathesis.

Atopic diathesis: asthma, hay fever and food allergies

Asthma is a chronic inflammatory disease of the lower respiratory tract caused by inflammation and remodeling of the airways. The inflammation is Th2 driven and IL-4 and IL-13 play the primary roles.[59,60] Asthma usually presents with coughing, wheezing and shortness of breath and is characterized by expiratory flow restriction due to bronchial hyperresponsiveness. The symptoms can be triggered by non-specific stimuli such as smoke and cold but reactions and sensitization to aeroallergens are found frequently.[61] The incidence is rising and 75% of the patients develop their asthma before the age of 7.[62] Having had AD as an infant increases the risk of developing asthma by two-fold (table 3).[42] About 40% of children with AD with IgE-sensitization will develop asthma.[63,64] Patients with early onset AD, multiple sensitizations and familial history of asthma are at highest risk of developing asthma.[65,66]

Table 3. Odds ratios and chances of asthma, hayfever and food allergies in atopic dermatitis

Atopic disorder	Fold increase of chance in AD	Chance in AD
Asthma	2	Mild AD: 20% Severe AD: 60%
Hayfever	2	50%
Food allergies	3-6	Mild AD: 5% Severe AD: 30%

Hay fever consist of the syndromes of allergic rhinitis and allergic conjunctivitis and presents with itchy, red eyes, sneezing and nasal congestion. Its prevalence is approximated between 10 and 30% in the industrialized countries.[67] Roughly 50% of children with AD develop hay fever.[63] Sensitization to common allergens can be identified through serum IgE testing or skin prick tests. However, the diagnosis can be made on the basis of clinical findings only.

Cumulative incidence of food allergy is around 5% by the age of 2 years.[68] IgE-mediated food allergies typically present with urticarial, oral and gastrointestinal symptoms, dyspnea and wheezing and anaphylaxis. Although late-onset reactions have been reported aggravation of AD by food allergies is debatable.[69] Allergies to cow's milk, hen's egg and wheat usually resolve after infancy but those to peanuts and fish typical persist and exhibit more severe symptoms.[69] Sensitization to food allergens can be found in up to two thirds of infants with moderate to severe AD and food allergies can be found in up to 30% of severe infant cases of AD.[70] The prevalence in mild cases and in adults is much lower.[71,72]

Non-atopic comorbidities

The association between atopic disorders as a group has been known for many years. Therefore, atopy is considered a spectrum of diseases or a diathesis. More recently, associations with other, non-atopic diseases are increasingly recognized. Atopic disorders have been linked to a number of Mendelian inherited disorders. These are ichtyosis vulgaris, Netherton's syndrome, Job's syndrome, DiGeorge syndrome, Nezelof syndrome, selective IgA deficiency, Wiskott-Aldrich syndrome, peeling skin syndrome type B and skin dermatitis-multiple severe allergies-metabolic wasting syndrome .[73] Studying these comorbidities has provided insight into the pathophysiology of atopic disorders.[74]

However, the list of co-morbidities not associated with single gene mutations is even longer.

Every year new comorbidities are added to it, some of which are considered to be truly a part of the atopic constitution. This is the case with eosinophilic esophagitis for example. The studies which have specifically and directly assessed the association between AD

and a possible comorbidity are listed in (Table 4). It is worth noting that the inflammatory and infectious disorders mainly colocalize in the skin.

Table 4. Non-atopic comorbidities associated with atopic dermatitis and atopy in general

Atopic condition	Comorbidity	Highest level of evidence studies
Atopic dermatitis	**Inflammatory**	
	Alopecia areata	Meta-analysis[70]
	Vitiligo	Meta-analysis[70]
	Lichen sclerosus	Case-control[71]
	Atopic eruption of pregnancy	Case series[72]
	Infectious	
	Eczema herpeticum	Case series[73]
	Verruca vulgaris	Cohort[74]
	Molluscum contagiosum	Review[75]
	Impetigo	Case-control[76]
	Upper and lower airway infections Gianotti-Crosti syndrome	Cohort[74] Case-control[77,78]
	Neoplastic	
	Skin cancer	Cohort [79-81]
	Lymphoma **Psychiatric**	Systematic review[82]
	Attention deficit hyperactivity disorder	Meta-analysis[83]
	Depression	Cohort[84]
	Autism	Systematic review[85]
	Cardio vascular	
	Obesity	Meta-analysis[86]
	Hypertension	Cohort[87]
	Myocardial infarction	Cohort[87]
	Stroke	Cohort[88]
General atopy	**Inflammatory**	
	Keratoconus	Case control[89]
	Retinal detachment	Cohort[90]
	Eosinophilic esophagitis	Review[91]
	Cataract	Case series[92]
	Glaucoma	Case series[93]
	Infectious HIV Respiratory viral infections **Neoplastic**	Review[94] Meta-analysis[95]
	Keloid	Case series[96]

Possible biologic pathways in comorbid disease

Several of biologic mechanisms could explain how impediments on the molecular level in AD possibly contribute to the comorbidities found in epidemiologic studies. First, the defective epidermal barrier allows penetration by damaging agents such as toxins, radiation and microorganisms. This obviously predisposes to infections and inflammatory disorders. Second, the Th-2 polarized immune system fails to address intracellular pathogens leading to infections.[102] There is further an increased turnover of epithelial and hematopoietic cells which could lead to mutations and consequently cancer. The continuous state of inflammation may also reflect on the cardiovascular system through increased circulating levels of pro-inflammatory molecules i.e. systemic inflammation. This is illustrated in (Figure 4).

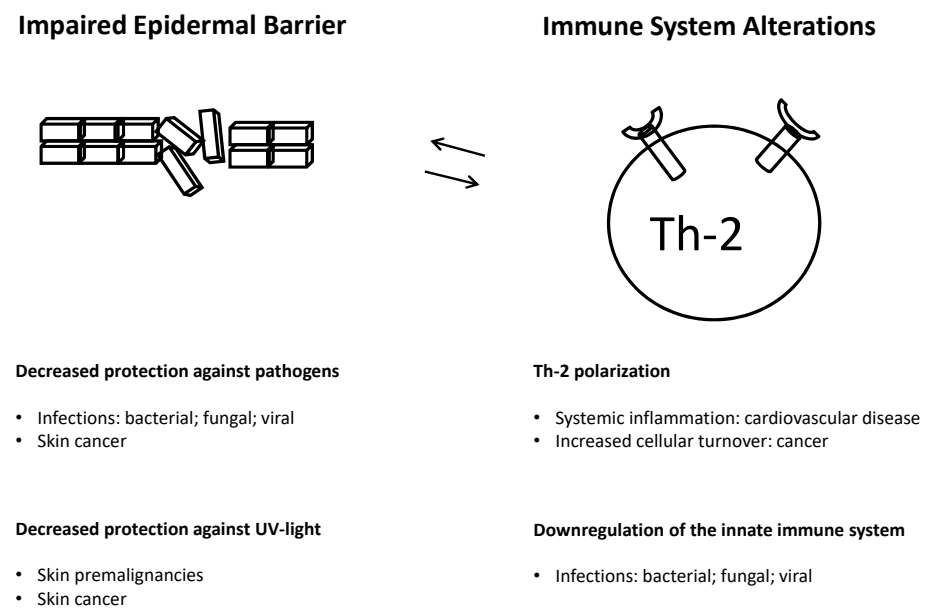

Figure 4. Hypothetical comorbidity pathways in atopic dermatitis

Anti-morbidities

Aside from the research performed in the field of the comorbidities of atopy almost equal attention has been given to the phenomenon of the decreased risk of auto-immune and auto-inflammatory disorders in atopic individuals.[103] Various epidemiologic studies have shown a reduced prevalence of atopic disorders in type 1 diabetes and rheumatoid arthritis (RA).[104-112] There is also some evidence for this phenomenon in multiple sclerosis.[113,114] The conceptual framework for these findings has traditionally been provided by the paradigm of the T-helper subsets and their mutual antagonism (figure 5).

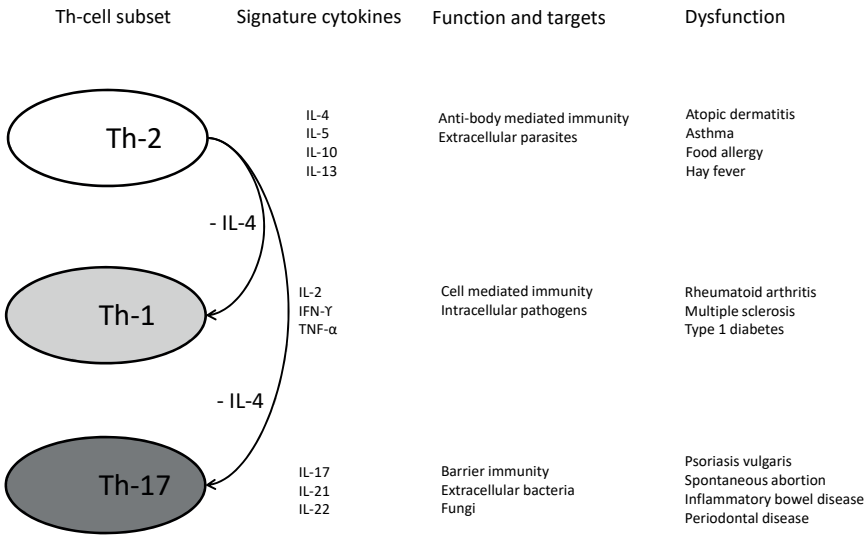

Figure 5. T-helper lymphocyte subsets and their functions.

The known types of inflammation are categorized according to the main T-helper cell subset involved. From vast quantities of research it is clear that during certain phases of inflammation, specific T-helper cells and their cytokines predominate in affected tissues. However, the Th-cells seem to be able to switch roles and to relinquish their dominance. This is seen in the chronic phases of AD and asthma where Th-17 cells take over from Th-2 cells. Moreover, two different lineages can be at work at the same time as Th-1 and Th-17 axes seem to play an equal role in rheumatoid arthritis. It is therefore not possible to categorize all inflammatory disorders in static divisions such as presented in figure 3 which is an illustration of a heuristic approach. In this figure the novel discovery of Th-22 and Th-9 cells has been left out as their pleiotropical functions have not yet been fully characterized.[115-118] To complicate matters even more, there is in vivo and in vitro evidence that the Th-cell subsets antagonize each other's functions. Th1/Th17 and Th2 types of inflammation control exhibit opposing immunologic mechanisms and the signature cytokines of Th-1 cells can make the Th-2 cells switch class.[119-121] On the other hand IL-4 ameliorates the inflammatory effects of Th-1 cells while IFN-γ and IL-4 inhibit Th-17 differentiation.[122] Summarizing, some auto-inflammatory and auto-immune disorders may be either precluded or attenuated by Th-2 polarized atopy. This leads to the inverse associations between these disorders and atopy. They could be perceived as contraries of comorbidities: anti-morbidities.

AIMS OF THIS THESIS

The main objective of this thesis was to explore and assess the associations of non-atopic comorbidities with atopic disorders and how atopy influences the course and/or severity of these conditions. The comorbidities are divided into inflammatory, cancerous and cardiovascular disorders. The chapters of this thesis correspond to this division.

In the first part of the **second chapter** we have assessed for associations between atopy and psoriasis vulgaris and psoriatic arthritis. Psoriasis vulgaris is a prevalent inflammatory skin disease which has psoriatic arthritis as its commonest comorbidity. They share the immunopathogenetic pathways which are mainly Th-17 oriented. A possible positive association with atopy would be of great interest as psoriasis vulgaris is also thought to stem from epidermal barrier dysfunction. A negative association could be expected as well because of a possible Th2-Th17 antagonism. In the second part of chapter 2 we describe the abnormalities seen within B-cells of patients with atopic dermatitis. The research on inflammatory skin disorders and particularly the research on AD has mainly focused on the T-lymphocytes. With IgE being the hall mark of atopy looking into the immunoglobulin producing B cells cannot be omitted.

The first part of **chapter 3** summarizes the evidence for the role of the epidermal barrier in the protection against UV-light induced cancers. Damage to this barrier could make humans more susceptible to keratinocyte cancers. The second part goes deeper into the association of skin cancer in AD in community dwelling patients. The third part assesses the association between melanoma and AD. With melanoma having the highest morbidity and mortality rates this was an obvious target for our research. The fundamental motive for this research are the phenomena of a damaged barrier (decrease in innate immunity) and Th-2 skewing (decreased tumor clearance by the Th-1 axis) which could lead to increased incidence of skin cancer in atopic individuals.

In **chapter 4** we have focused on the association between AD and cardiovascular disease (CVD) and cardiovascular risk factors. CVDs are increasingly the subject of dermatologic studies. Our study was done within the Rotterdam Study as this cohort was designed to deliver high quality, validated data on cardiovascular disease.

Chapter 5 is the general discussion commenting on the findings, the methodologies and their limitations and putting them in a broader scientific and clinical context.

REFERENCES

1. Ring J. Terminology of allergic phenomena. Chem Immunol Allergy 2014;100:46-52.
2. Stanworth DR. The "discovery" of IgE. Allergol Immunopathol (Madr) 1987;15:175-7.
3. Wuthrich B, Schmid-Grendelmeier P. The atopic eczema/dermatitis syndrome. Epidemiology, natural course, and immunology of the IgE-associated ("extrinsic") and the nonallergic ("intrinsic") AEDS. J Investig Allergol Clin Immunol 2003;13:1-5.
4. Weidinger S, Novak N. Atopic dermatitis. Lancet 2016;387:1109-22.
5. DaVeiga SP. Epidemiology of atopic dermatitis: a review. Allergy Asthma Proc 2012;33:227-34.
6. Williams H, Flohr C. How epidemiology has challenged 3 prevailing concepts about atopic dermatitis. J Allergy Clin Immunol 2006;118:209-13.
7. Pyun BY. Natural history and risk factors of atopic dermatitis in children. Allergy Asthma Immunol Res 2015;7:101-5.
8. Nutten S. Atopic dermatitis: global epidemiology and risk factors. Ann Nutr Metab 2015;66 Suppl 1:8-16.
9. Czarnowicki T, Krueger JG, Guttman-Yassky E. Skin barrier and immune dysregulation in atopic dermatitis: an evolving story with important clinical implications. J Allergy Clin Immunol Pract 2014;2:371-9; quiz 80-1.
10. Palmer CN, Irvine AD, Terron-Kwiatkowski A, et al. Common loss-of-function variants of the epidermal barrier protein filaggrin are a major predisposing factor for atopic dermatitis. Nat Genet 2006;38:441-6.
11. Weidinger S, Illig T, Baurecht H, et al. Loss-of-function variations within the filaggrin gene predispose for atopic dermatitis with allergic sensitizations. J Allergy Clin Immunol 2006;118:214-9.
12. Sandilands A, Terron-Kwiatkowski A, Hull PR, et al. Comprehensive analysis of the gene encoding filaggrin uncovers prevalent and rare mutations in ichthyosis vulgaris and atopic eczema. Nat Genet 2007;39:650-4.
13. Sandilands A, Sutherland C, Irvine AD, McLean WH. Filaggrin in the frontline: role in skin barrier function and disease. Journal of cell science 2009;122:1285-94.
14. Denecker G, Ovaere P, Vandenabeele P, Declercq W. Caspase-14 reveals its secrets. The Journal of cell biology 2008;180:451-8.
15. Hoste E, Kemperman P, Devos M, et al. Caspase-14 is required for filaggrin degradation to natural moisturizing factors in the skin. J Invest Dermatol 2011;131:2233-41.
16. Eckhart L, Tschachler E. Cuts by caspase-14 control the proteolysis of filaggrin. J Invest Dermatol 2011;131:2173-5.
17. Barresi C, Stremnitzer C, Mlitz V, et al. Increased sensitivity of histidinemic mice to UVB radiation suggests a crucial role of endogenous urocanic acid in photoprotection. J Invest Dermatol 2011;131:188-94.
18. Gibbs NK, Tye J, Norval M. Recent advances in urocanic acid photochemistry, photobiology and photoimmunology. Photochem Photobiol Sci 2008;7:655-67.
19. Vasilopoulos Y, Cork MJ, Murphy R, et al. Genetic association between an AACC insertion in the 3'UTR of the stratum corneum chymotryptic enzyme gene and atopic dermatitis. J Invest Dermatol 2004;123:62-6.
20. Soderhall C, Marenholz I, Kerscher T, et al. Variants in a novel epidermal collagen gene (COL29A1) are associated with atopic dermatitis. PLoS Biol 2007;5:e242.

21. Engebretsen KA, Linneberg A, Thuesen BH, et al. Xerosis is associated with asthma in men independent of atopic dermatitis and filaggrin gene mutations. Journal of the European Academy of Dermatology and Venereology : JEADV 2015;29:1807-15.
22. Boguniewicz M, Leung DY. Atopic dermatitis: a disease of altered skin barrier and immune dysregulation. Immunological reviews 2011;242:233-46.
23. Le Lamer M, Pellerin L, Reynier M, et al. Defects of corneocyte structural proteins and epidermal barrier in atopic dermatitis. Biol Chem 2015;396:1163-79.
24. Brown SJ, McLean WH. One remarkable molecule: filaggrin. J Invest Dermatol 2012;132:751-62.
25. Henderson J, Northstone K, Lee SP, et al. The burden of disease associated with filaggrin mutations: a population-based, longitudinal birth cohort study. J Allergy Clin Immunol 2008;121:872-7 e9.
26. Marenholz I, Nickel R, Ruschendorf F, et al. Filaggrin loss-of-function mutations predispose to phenotypes involved in the atopic march. J Allergy Clin Immunol 2006;118:866-71.
27. Suarez-Farinas M, Tintle SJ, Shemer A, et al. Nonlesional atopic dermatitis skin is characterized by broad terminal differentiation defects and variable immune abnormalities. J Allergy Clin Immunol 2011;127:954-64 e1-4.
28. Bonnelykke K, Sparks R, Waage J, Milner JD. Genetics of allergy and allergic sensitization: common variants, rare mutations. Current opinion in immunology 2015;36:115-26.
29. Mu Z, Zhao Y, Liu X, Chang C, Zhang J. Molecular biology of atopic dermatitis. Clin Rev Allergy Immunol 2014;47:193-218.
30. Doherty TA. At the bench: understanding group 2 innate lymphoid cells in disease. J Leukoc Biol 2015;97:455-67.
31. Cardona ID, Cho SH, Leung DY. Role of bacterial superantigens in atopic dermatitis : implications for future therapeutic strategies. Am J Clin Dermatol 2006;7:273-9.
32. Nakatsuji T, Chen TH, Two AM, et al. Staphylococcus aureus exploits epidermal barrier defects in atopic dermatitis to trigger cytokine expression. J Invest Dermatol 2016.
33. Li Z, Levast B, Madrenas J. Staphylococcus aureus Downregulates IP-10 Production and Prevents Th1 Cell Recruitment. J Immunol 2017.
34. Bunikowski R, Mielke M, Skarabis H, et al. Prevalence and role of serum IgE antibodies to the Staphylococcus aureus-derived superantigens SEA and SEB in children with atopic dermatitis. J Allergy Clin Immunol 1999;103:119-24.
35. Hepburn L, Hijnen DJ, Sellman BR, et al. The complex biology and contribution of Staphylococcus aureus in atopic dermatitis, current and future therapies. Br J Dermatol 2016.
36. Flohr C, Johansson SG, Wahlgren CF, Williams H. How atopic is atopic dermatitis? J Allergy Clin Immunol 2004;114:150-8.
37. Kerzel S, Rogosch T, Struecker B, Maier RF, Kabesch M, Zemlin M. Unlike in Children with Allergic Asthma, IgE Transcripts from Preschool Children with Atopic Dermatitis Display Signs of Superantigen-Driven Activation. J Immunol 2016.
38. Tang TS, Bieber T, Williams HC. Does "autoreactivity" play a role in atopic dermatitis? J Allergy Clin Immunol 2012;129:1209-15 e2.
39. Thijs J, Krastev T, Weidinger S, et al. Biomarkers for atopic dermatitis: a systematic review and meta-analysis. Curr Opin Allergy Clin Immunol 2015;15:453-60.
40. Thijs JL, van Seggelen W, Bruijnzeel-Koomen C, de Bruin-Weller M, Hijnen D. New Developments in Biomarkers for Atopic Dermatitis. J Clin Med 2015;4:479-87.
41. Tsybikov NN, Petrisheva IV, Fefelova EV, Kuznik BI, Magen E. Plasma alpha-defensins are elevated during exacerbation of atopic dermatitis. Clin Exp Dermatol 2016;41:253-9.

42. Dharmage SC, Lowe AJ, Matheson MC, Burgess JA, Allen KJ, Abramson MJ. Atopic dermatitis and the atopic march revisited. Allergy 2014;69:17-27.
43. Mortz CG, Andersen KE, Dellgren C, Barington T, Bindslev-Jensen C. Atopic dermatitis from adolescence to adulthood in the TOACS cohort: prevalence, persistence and comorbidities. Allergy 2015;70:836-45.
44. Jacob SE, Goldenberg A, Nedorost S, Thyssen JP, Fonacier L, Spiewak R. Flexural eczema versus atopic dermatitis. Dermatitis 2015;26:109-15.
45. Saeki H, Nakahara T, Tanaka A, et al. Clinical Practice Guidelines for the Management of Atopic Dermatitis 2016. J Dermatol 2016.
46. Eichenfield LF, Tom WL, Berger TG, et al. Guidelines of care for the management of atopic dermatitis: section 2. Management and treatment of atopic dermatitis with topical therapies. J Am Acad Dermatol 2014;71:116-32.
47. Broeders JA, Ahmed Ali U, Fischer G. Systematic review and meta-analysis of randomized clinical trials (RCTs) comparing topical calcineurin inhibitors with topical corticosteroids for atopic dermatitis: A 15-year experience. J Am Acad Dermatol 2016.
48. Chia BK, Tey HL. Systematic review on the efficacy, safety, and cost-effectiveness of topical calcineurin inhibitors in atopic dermatitis. Dermatitis 2015;26:122-32.
49. Hanifin JM, Ellis CN, Frieden IJ, et al. OPA-15406, a novel, topical, nonsteroidal, selective phosphodiesterase-4 (PDE4) inhibitor, in the treatment of adult and adolescent patients with mild to moderate atopic dermatitis (AD): A phase-II randomized, double-blind, placebo-controlled study. J Am Acad Dermatol 2016.
50. Sidbury R, Davis DM, Cohen DE, et al. Guidelines of care for the management of atopic dermatitis: section 3. Management and treatment with phototherapy and systemic agents. J Am Acad Dermatol 2014;71:327-49.
51. Hon KL, Ching GK, Leung TF, Chow CM, Lee KK, Ng PC. Efficacy and tolerability at 3 and 6 months following use of azathioprine for recalcitrant atopic dermatitis in children and young adults. J Dermatolog Treat 2009;20:141-5.
52. Schmitt J, Schakel K, Folster-Holst R, et al. Prednisolone vs. ciclosporin for severe adult eczema. An investigator-initiated double-blind placebo-controlled multicentre trial. Br J Dermatol 2010;162:661-8.
53. Haeck IM, Knol MJ, Ten Berge O, van Velsen SG, de Bruin-Weller MS, Bruijnzeel-Koomen CA. Enteric-coated mycophenolate sodium versus cyclosporin A as long-term treatment in adult patients with severe atopic dermatitis: a randomized controlled trial. J Am Acad Dermatol 2011;64:1074-84.
54. Schram ME, Roekevisch E, Leeflang MM, Bos JD, Schmitt J, Spuls PI. A randomized trial of methotrexate versus azathioprine for severe atopic eczema. J Allergy Clin Immunol 2011;128:353-9.
55. Thaci D, Simpson EL, Beck LA, et al. Efficacy and safety of dupilumab in adults with moderate-to-severe atopic dermatitis inadequately controlled by topical treatments: a randomised, placebo-controlled, dose-ranging phase 2b trial. Lancet 2016;387:40-52.
56. Beck LA, Thaci D, Hamilton JD, et al. Dupilumab treatment in adults with moderate-to-severe atopic dermatitis. N Engl J Med 2014;371:130-9.
57. Sidbury R, Tom WL, Bergman JN, et al. Guidelines of care for the management of atopic dermatitis: Section 4. Prevention of disease flares and use of adjunctive therapies and approaches. J Am Acad Dermatol 2014;71:1218-33.
58. Valderas JM, Starfield B, Sibbald B, Salisbury C, Roland M. Defining comorbidity: implications for understanding health and health services. Annals of family medicine 2009;7:357-63.

59. Postma DS, Rabe KF. The Asthma-COPD Overlap Syndrome. N Engl J Med 2015;373:1241-9.
60. Postma DS, Kerstjens HA. Characteristics of airway hyperresponsiveness in asthma and chronic obstructive pulmonary disease. Am J Respir Crit Care Med 1998;158:S187-92.
61. Bateman ED, Hurd SS, Barnes PJ, et al. Global strategy for asthma management and prevention: GINA executive summary. Eur Respir J 2008;31:143-78.
62. Yunginger JW, Reed CE, O'Connell EJ, Melton LJ, 3rd, O'Fallon WM, Silverstein MD. A community-based study of the epidemiology of asthma. Incidence rates, 1964-1983. Am Rev Respir Dis 1992;146:888-94.
63. Gustafsson D, Sjoberg O, Foucard T. Development of allergies and asthma in infants and young children with atopic dermatitis--a prospective follow-up to 7 years of age. Allergy 2000;55:240-5.
64. Spergel JM. Epidemiology of atopic dermatitis and atopic march in children. Immunol Allergy Clin North Am 2010;30:269-80.
65. Amat F, Saint-Pierre P, Bourrat E, et al. Early-onset atopic dermatitis in children: which are the phenotypes at risk of asthma? Results from the ORCA cohort. PLoS One 2015;10:e0131369.
66. Spergel JM, Paller AS. Atopic dermatitis and the atopic march. J Allergy Clin Immunol 2003;112:S118-27.
67. Singh K, Axelrod S, Bielory L. The epidemiology of ocular and nasal allergy in the United States, 1988-1994. J Allergy Clin Immunol 2010;126:778-83 e6.
68. Grimshaw KE, Bryant T, Oliver EM, et al. Incidence and risk factors for food hypersensitivity in UK infants: results from a birth cohort study. Clin Transl Allergy 2015;6:1.
69. Longo G, Berti I, Burks AW, Krauss B, Barbi E. IgE-mediated food allergy in children. Lancet 2013;382:1656-64.
70. Eller E, Kjaer HF, Host A, Andersen KE, Bindslev-Jensen C. Food allergy and food sensitization in early childhood: results from the DARC cohort. Allergy 2009;64:1023-9.
71. Worm M, Forschner K, Lee HH, et al. Frequency of atopic dermatitis and relevance of food allergy in adults in Germany. Acta Derm Venereol 2006;86:119-22.
72. Martin PE, Eckert JK, Koplin JJ, et al. Which infants with eczema are at risk of food allergy? Results from a population-based cohort. Clin Exp Allergy 2015;45:255-64.
73. Cookson WO, Moffatt MF. Genetics of asthma and allergic disease. Hum Mol Genet 2000;9:2359-64.
74. Samuelov L, Sprecher E. Peeling off the genetics of atopic dermatitis-like congenital disorders. J Allergy Clin Immunol 2014;134:808-15.
75. Mohan GC, Silverberg JI. Association of Vitiligo and Alopecia Areata With Atopic Dermatitis: A Systematic Review and Meta-analysis. JAMA dermatology 2015;151:522-8.
76. Becker K, Meissner V, Farwick W, Bauer R, Gaiser MR. Lichen sclerosus and atopy in boys: coincidence or correlation? Br J Dermatol 2013;168:362-6.
77. Roth MM, Cristodor P, Kroumpouzos G. Prurigo, pruritic folliculitis, and atopic eruption of pregnancy: Facts and controversies. Clinics in dermatology 2016;34:392-400.
78. Bork K, Brauninger W. Increasing incidence of eczema herpeticum: analysis of seventy-five cases. J Am Acad Dermatol 1988;19:1024-9.
79. Silverberg JI, Silverberg NB. Childhood atopic dermatitis and warts are associated with increased risk of infection: a US population-based study. J Allergy Clin Immunol 2014;133:1041-7.
80. Olsen JR, Gallacher J, Piguet V, Francis NA. Epidemiology of molluscum contagiosum in children: a systematic review. Family practice 2014;31:130-6.

81. Hayashida S, Furusho N, Uchi H, et al. Are lifetime prevalence of impetigo, molluscum and herpes infection really increased in children having atopic dermatitis? Journal of dermatological science 2010;60:173-8.
82. Ricci G, Patrizi A, Neri I, Specchia F, Tosti G, Masi M. Gianotti-Crosti syndrome and allergic background. Acta Derm Venereol 2003;83:202-5.
83. Chuh A, Zawar V, Lee A, Sciallis G. Is Gianotti-Crosti Syndrome Associated with Atopy? A Case-Control Study and a Postulation on the Intrinsic Host Factors in Gianotti-Crosti Syndrome. Pediatric dermatology 2016.
84. Dyer RK, Weinstock MA, Cohen TS, Rizzo AE, Bingham SF, Group VT. Predictors of basal cell carcinoma in high-risk patients in the VATTC (VA Topical Tretinoin Chemoprevention) trial. J Invest Dermatol 2012;132:2544-51.
85. Arana A, Wentworth CE, Fernandez-Vidaurre C, Schlienger RG, Conde E, Arellano FM. Incidence of cancer in the general population and in patients with or without atopic dermatitis in the U.K. Br J Dermatol 2010;163:1036-43.
86. Jensen AO, Svaerke C, Kormendine Farkas D, Olesen AB, Kragballe K, Sorensen HT. Atopic dermatitis and risk of skin cancer: a Danish nationwide cohort study (1977-2006). Am J Clin Dermatol 2012;13:29-36.
87. Legendre L, Barnetche T, Mazereeuw-Hautier J, Meyer N, Murrell D, Paul C. Risk of lymphoma in patients with atopic dermatitis and the role of topical treatment: A systematic review and meta-analysis. J Am Acad Dermatol 2015;72:992-1002.
88. Schmitt J, Apfelbacher C, Heinrich J, Weidinger S, Romanos M. [Association of atopic eczema and attention-deficit/hyperactivity disorder - meta-analysis of epidemiologic studies]. Zeitschrift fur Kinder- und Jugendpsychiatrie und Psychotherapie 2013;41:35-42; quiz -4.
89. Yu SH, Silverberg JI. Association between Atopic Dermatitis and Depression in US Adults. J Invest Dermatol 2015;135:3183-6.
90. Billeci L, Tonacci A, Tartarisco G, Ruta L, Pioggia G, Gangemi S. Association Between Atopic Dermatitis and Autism Spectrum Disorders: A Systematic Review. Am J Clin Dermatol 2015;16:371-88.
91. Zhang A, Silverberg JI. Association of atopic dermatitis with being overweight and obese: a systematic review and metaanalysis. J Am Acad Dermatol 2015;72:606-16 e4.
92. Silverberg JI, Greenland P. Eczema and cardiovascular risk factors in 2 US adult population studies. J Allergy Clin Immunol 2015.
93. Su VY, Chen TJ, Yeh CM, et al. Atopic dermatitis and risk of ischemic stroke: a nationwide population-based study. Ann Med 2014;46:84-9.
94. Bawazeer AM, Hodge WG, Lorimer B. Atopy and keratoconus: a multivariate analysis. The British journal of ophthalmology 2000;84:834-6.
95. Hida T, Tano Y, Okinami S, Ogino N, Inoue M. Multicenter retrospective study of retinal detachment associated with atopic dermatitis. Japanese journal of ophthalmology 2000;44:407-18.
96. Noel RJ, Putnam PE, Rothenberg ME. Eosinophilic esophagitis. N Engl J Med 2004;351:940-1.
97. Bair B, Dodd J, Heidelberg K, Krach K. Cataracts in atopic dermatitis: a case presentation and review of the literature. Arch Dermatol 2011;147:585-8.
98. Takakuwa K, Hamanaka T, Mori K, et al. Atopic Glaucoma: Clinical and Pathophysiological Analysis. Journal of glaucoma 2015;24:662-8.
99. Brennan C. Up to 70% of HIV-positive individuals report experiencing symptoms related to atopy. HIV clinician / Delta Region AIDS Education & Training Center 2015;27:1-10.

100. Shi T, Balsells E, Wastnedge E, et al. Risk factors for respiratory syncytial virus associated with acute lower respiratory infection in children under five years: Systematic review and meta-analysis. Journal of global health 2015;5:020416.
101. Smith CJ, Smith JC, Finn MC. The possible role of mast cells (allergy) in the production of keloid and hypertrophic scarring. The Journal of burn care & rehabilitation 1987;8:126-31.
102. Hajdarbegovic E, Verkouteren J, Balak D. Non-melanoma skin cancer: the hygiene hypothesis. Medical hypotheses 2012;79:872-4.
103. Rabin RL, Levinson AI. The nexus between atopic disease and autoimmunity: a review of the epidemiological and mechanistic literature. Clinical and experimental immunology 2008;153:19-30.
104. Stromberg LG, Ludvigsson GJ, Bjorksten B. Atopic allergy and delayed hypersensitivity in children with diabetes. J Allergy Clin Immunol 1995;96:188-92.
105. Decreased prevalence of atopic diseases in children with diabetes. The EURODIAB Substudy 2 Study Group. The Journal of pediatrics 2000;137:470-4.
106. Stene LC, Ronningen KS, Bjornvold M, Undlien DE, Joner G. An inverse association between history of childhood eczema and subsequent risk of type 1 diabetes that is not likely to be explained by HLA-DQ, PTPN22, or CTLA4 polymorphisms. Pediatric diabetes 2010;11:386-93.
107. Rosenbauer J, Herzig P, Giani G. Atopic eczema in early childhood could be protective against Type 1 diabetes. Diabetologia 2003;46:784-8.
108. Douek IF, Leech NJ, Bingley PJ, Gale EA. Eczema and Type 1 diabetes. Diabetic medicine : a journal of the British Diabetic Association 2002;19:174-5.
109. Verhoef CM, van Roon JA, Vianen ME, Bruijnzeel-Koomen CA, Lafeber FP, Bijlsma JW. Mutual antagonism of rheumatoid arthritis and hay fever; a role for type 1/type 2 T cell balance. Annals of the rheumatic diseases 1998;57:275-80.
110. Rudwaleit M, Andermann B, Alten R, et al. Atopic disorders in ankylosing spondylitis and rheumatoid arthritis. Annals of the rheumatic diseases 2002;61:968-74.
111. Hilliquin P, Allanore Y, Coste J, Renoux M, Kahan A, Menkes CJ. Reduced incidence and prevalence of atopy in rheumatoid arthritis. Results of a case-control study. Rheumatology 2000;39:1020-6.
112. Olesen AB, Juul S, Birkebaek N, Thestrup-Pedersen K. Association between atopic dermatitis and insulin-dependent diabetes mellitus: a case-control study. Lancet 2001;357:1749-52.
113. Oro AS, Guarino TJ, Driver R, Steinman L, Umetsu DT. Regulation of disease susceptibility: decreased prevalence of IgE-mediated allergic disease in patients with multiple sclerosis. J Allergy Clin Immunol 1996;97:1402-8.
114. Tremlett HL, Evans J, Wiles CM, Luscombe DK. Asthma and multiple sclerosis: an inverse association in a case-control general practice population. QJM : monthly journal of the Association of Physicians 2002;95:753-6.
115. Deng Y, Wang Z, Chang C, Lu L, Lau CS, Lu Q. Th9 cells and IL-9 in autoimmune disorders: Pathogenesis and therapeutic potentials. Hum Immunol 2016.
116. Clark RA, Schlapbach C. TH9 cells in skin disorders. Semin Immunopathol 2017;39:47-54.
117. Azizi G, Simhag A, El Rouby NM, Mirshafiey A. Th22 Cells Contribution in Immunopathogenesis of Rheumatic Diseases. Iran J Allergy Asthma Immunol 2015;14:246-54.
118. Eyerich K, Eyerich S. Th22 cells in allergic disease. Allergo J Int 2015;24:1-7.
119. Sornasse T, Larenas PV, Davis KA, de Vries JE, Yssel H. Differentiation and stability of T helper 1 and 2 cells derived from naive human neonatal CD4+ T cells, analyzed at the single-cell level. The Journal of experimental medicine 1996;184:473-83.

120. Baurecht H, Hotze M, Brand S, et al. Genome-wide comparative analysis of atopic dermatitis and psoriasis gives insight into opposing genetic mechanisms. American journal of human genetics 2015;96:104-20.
121. Eyerich S, Onken AT, Weidinger S, et al. Mutual antagonism of T cells causing psoriasis and atopic eczema. N Engl J Med 2011;365:231-8.
122. Tesmer LA, Lundy SK, Sarkar S, Fox DA. Th17 cells in human disease. Immunological reviews 2008;223:87-113.

Chapter 2

Atopy and immune disorders

Chapter 2.1

Decreased prevalence of atopic features in patients with psoriatic arthritis, but not in psoriasis vulgaris

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ABSTRACT

Background: The prevalence of atopic disorders is reduced in patients with various autoinflammatory diseases, but this association has not been studied in psoriasis vulgaris or psoriatic arthritis.

Objective: To compare the prevalence of hayfever, asthma and sensitization to common aeroallergens in patients with psoriasis vulgaris and patients with psoriatic arthritis to controls and to investigate whether atopy influences the arthritis activity and severity scores in psoriatic arthritis patients.

Methods: In a cross-sectional cohort study design, the differences in patient-reported lifetime prevalence of atopic disorders and serum immunoglobulin E directed against common aeroallergens were compared. The effect of atopy on arthritis severity was assessed using the Disease Activity Score 28 and Health Assessment Questionnaire. Logistic regression models were used to calculate crude and adjusted odds ratios with 95%-confidence intervals for presence of atopy.

Results: One hundred and sixty-eight patients with psoriatic arthritis, 133 psoriasis vulgaris patients and 147 controls were included. The life time prevalence of hayfever did not differ across groups. Patients with psoriatic arthritis were less likely to have suffered from asthma than controls; adjusted OR=0.20 [CI 95% 0.04-0.92] and they were less likely to be sensitized; adjusted OR=0.50 [95%CI 0.25-0.99]. HAQ-VAS for pain and HAQ-VAS for patient global score were significantly reduced by sensitization to common aeroallergens (beta-coefficient -0.54 [95% CI -0.84 - -0.25] and -18.4 [95% CI -28.5 - -8.25] respectively.)

Limitations : This is a cross-sectional, small numbered study.

Conclusion: Atopy may protect against development of psoriatic arthritis and diminish its severity.

Key words

Atopy, asthma, hayfever, psoriatic arthritis, psoriasis vulgaris, life-time prevalence

Capsule summary

The prevalence of atopic disorders is diminished in rheumatoid arthritis.
The prevalence of atopic disorders is also diminished in patients with psoriatic arthritis.
Atopy may be a risk determinant and a severity predictor for psoriatic arthritis.

ABBREVIATIONS

PSO	psoriasis vulgaris or chronic plaque type psoriasis
PSA	psoriatic arthritis
OR	odds ratio
RA	rheumatoid arthritis
95% CI	95% confidence interval
Th	T-helper cell
CASPAR	Classification Criteria for Psoriatic Arthritis
PASI	Psoriasis Area and Severity Index
DAS 28	Disease Activity Score 28
HAQDI	Health Assessment Questionaire-Disability Index
HAQ-VAS	Health Assessment Questionnaire Visual Analogue Scale
SD	standard deviation

INTRODUCTION

Psoriasis vulgaris (PSO) is a common inflammatory skin disorder affecting around 2% of the Western population.[1] Approximately 11% (the estimates go up to 24%) of PSO patients also have psoriatic arthritis (PSA)[2]. It remains controversial whether PSO and PSA are part of the same disease process or merely embody a strong association between two distinct entities with overlap in genetic susceptibility loci.[3] Despite the enormous differences in clinical manifestations, the inflammation in both diseases is concerted by Th-17 (T-helper) and Th-1 cells.[4] While Th-17 and Th-1 cells in cutaneous and arthropatic psoriasis may work synergistically to produce the inflammation seen in these disorders, type-2 Th-cells have been shown to antagonize the inflammation caused by these cells on a cytokine level.[5-6]

Th-2 cells are important players in inflammation seen in atopic disorders.[7]

Atopy is a hereditary predisposition to allergic sensitization, i.e. development of IgE antibodies directed against various allergens. Clinically, it is a risk factor for the development of atopic dermatitis, hayfever and atopic asthma.

There is a multitude of epidemiologic studies which have shown lower prevalences of atopic disorders in patients with Th-1 mediated diseases. [8] These observational studies have shown that in rheumatoid arthritis (RA), which is a Th-1 driven disease, patients have lower prevalences of atopic disorders compared to controls. Moreover, hayfever may ameliorate the symptoms of RA.[9-11] These data suggest the existence of mutual exclusion or antagonism between atopic and autoinflammatory/autoimmune disorders, where Th-2 driven atopy protects against Th-1 driven disorders. The association between atopic disorders and PSA and the effect of atopy on PSA severity have not been studied yet.

The objective of this study was to compare the prevalence of clinical and serological manifestations of atopy in patients with PSA to PSO patients and healthy controls. We hypothesized that the prevalence of atopic disorders is lower in patients with PSO and PSA and that atopic PSA patients have a less severe disease

PATIENTS AND METHODS

Study population

The study subjects were recruited from March of 2009 until February of 2011. The PSA patients were recruited from the department of rheumatology at Maxima Medisch Centrum in Eindhoven. Patients with PSO and controls were recruited from the department of dermatology Erasmus Medical Centre in Rotterdam. All participating patients provided written informed consent. PSA diagnosis was confirmed by a certified rheumatologist and all patients fulfilled the CASPAR criteria[12]. All PSO subjects were diagnosed with psoriasis vulgaris by certified dermatologists and had no history or signs of inflammatory arthritis. The control group consisted of individuals with varicose veins who did not have either PSO or PSA. Only controls who had been referred for phlebological consultation by a general practitioner were included in order to avoid inclusion of atopic patients with concurrent varicosities referred by dermatologists.

Patient reported atopic disease

Directly after inclusion patients were given a questionnaire comprising questions from the European Community Respiratory Health Survey and International Study of Asthma and Allergies in Children protocol[13-14]. This questionnaire provided data on patient's self-reported symptomatology of atopic disorders (i.e. contact dermatitis, hayfever and asthma) during their life time. Subjects were considered to have asthma if they positively answered the question "Have you ever been diagnosed with asthma by a doctor?" and if the answer to question "How old were you when you were diagnosed with asthma?" was less than 25 years. For diagnosis of hayfever patients had to answer next question affirmatively; "Have you ever had hayfever?" and they would have to have it before the age of 30. We have left out atopic dermatitis from further analysis because of the clinical similarities with psoriasis vulgaris.

Total IgE levels and sensitization to common aeroallergens

As an objective marker of atopy IgE directed towards common aeroallergens together with total serum IgE was preferred to skin prick testing because of expenses and time consumption. Venous blood was drawn from all subjects and centrifuged to separate serum. All serum samples were frozen at -80 degrees Celsius and were later analyzed simultaneously as a batch. The sensitization analyses were performed at the Clinical Laboratory of the Erasmus Medical Center according to manufacturer's protocol (Phadia AB, Sweden). Levels of total IgE, and specific IgE directed to inhalant allergens (cat and dog dander, birch pollen, grass pollen, house dust mite and herb pollen) were determined. Levels of serum IgE higher than 100 kU/L were considered increased[15]. Patients were sensitized to aeroallergens if the serum value of IgE directed against aforementioned

allergens was more than 0.35 kU/L[16]. The results of total serum IgE analyses were not available for three PSO and 1 PSA patients because of loss of samples.

Arthritis severity

PSA patients were also given the Health Assessment Questionnaire [17] (HAQ), which produces a Disability Index (HAQ-DI) specific for arthritis patients as well as a visual analogue scale (VAS) for pain and patient global VAS. Clinical arthritis activity was assessed with Disease Activity Score for arthritis [18] (DAS28).

Statistics

For sample size calculations, we assumed a prevalence of 20% of atopic disorders in our control group[10] and expected half of this proportion in PSA and PSO patients. For a power of 80% and an alpha level of 0.05, a sample size of 135 cases and 135 controls was required. The demographic characteristics and frequency of the atopic features were statistically compared between three patient cohorts: PSO patients, PSA patients and a control group. The Chi-squared test was used to test for statistical differences in proportions and the Mann-Whitney U test was used to analyse the differences in nonparametric continuous variables (i.e. DAS28 and HAQ scores). In univariate logistic regression we have analyzed the effect of sex, age, methotrexate use, biological use and smoking on clinical and serological parameters of atopy. Variables with an effect with $p<0.20$ were included in the multivariate analyses. During preliminary analyses it was clear that the prevalence of atopic manifestations was not decreased in PSO patients. Therefore only patients with PSA were included in disease severity analyses. For the analysis of the effect of atopy on the severity of arthritis we have used sensitization to common allergens as definition for atopy. Sensitization to common allergens is more specific for atopy than total serum IgE and is more prevalent than clinical manifestations of atopy[19]. Variables were included in the multivariate models and linear regression analysis if $p<0.20$ in the univariate analysis. All data were analyzed using IBM SPSS version 19.0 (Chicago, Illinois). This study was approved by the medical ethical committees of Erasmus MC and Maxima Medical Centre (reference numbers MEC- 2007-181 and NL 1394307807) and was conducted in accordance with the Declaration of Helsinki.

RESULTS

Demographic characteristics

A total of 448 subjects were included of which 133 patients suffered from PSO, 168 with PSA and 147 were controls (Table 1). The mean age was comparable over the three groups and varied between 49 and 54 years. The proportion of males was highest in

the PSO group. On average, patients with PSO were younger than patients with PSA at onset of their disease. The PASI score was higher in PSO patients indicating more severe cutaneous disease compared with PSA. Patients with PSA were more likely to be on methotrexate (MTX) and as likely to be on a TNF-antagonist compared to the PSO population (Table 1).

Table 1. The distribution of patient and disease characteristics of the three patient cohorts

Group	PSO (n=133)	PSA (n=168)	Control* (n=147)
Mean age, years (SD)	49 (15.5)	52 (12.0)	54 (15.6)
Males (%)	60%	53%	44%
Age at onset in years (SD)	31 (17.4)	41 (13.8)	N/A
Duration of disease, years (SD)	30 (16.8)	11 (10.1)	N/A
PASI score (SD)	7.0 (6.8)	1.5 (2.5)	N/A
DAS28 score (SD)	N/A	2.6 (1.0)	N/A
Current MTX users	12%	49%	N/A
Current biological users	17%	14%	N/A
Current smokers	59%	27%	34%
Family history of atopy	28%	29%	27%

Abbreviations: PSO, psoriasis vulgaris; PSA, Psoriatic Arthritis, SD, Standard Deviation, PASI, Psoriasis Area and Severity Index (ranges from 0-72); DAS28, Disease Activity Score 28; MTX, methotrexate; N/A, not applicable

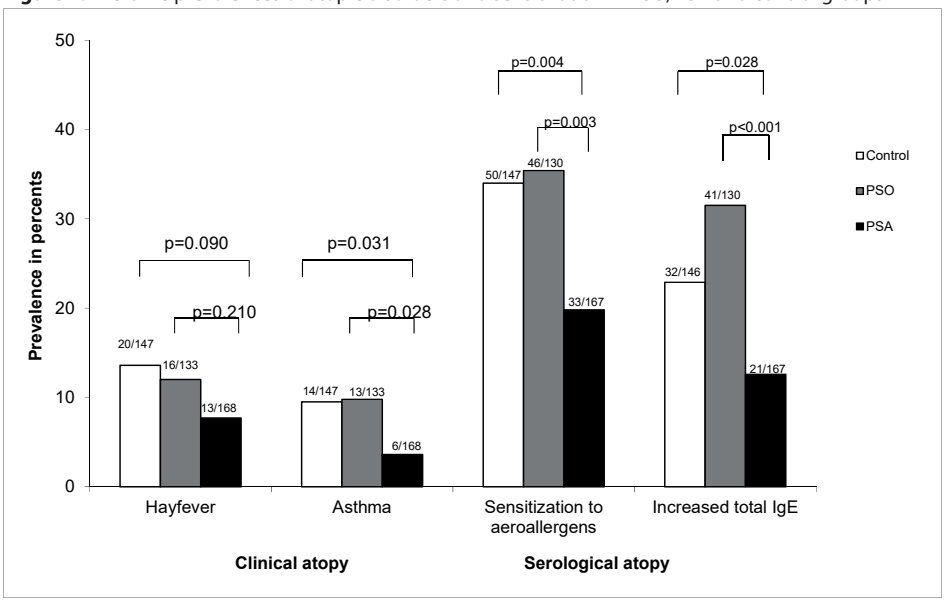
* patients referred to the department of dermatology for varicose veins.

Of the three patient groups, PSO patients reported significantly more frequently to be current smokers compared to PSA and control patients. A positive family history for atopic disorders was observed in almost 30% for each of the groups.

Hayfever

The proportion of subjects who reported having had hayfever was lower in the PSA group (7.7%) than in controls (13.6%) and in PSO group (12.0%), (p=0.09 and p=0.21 respectively: Figure 1). Age and methotrexate use potentially confounded the association between hayfever and PSA or PSO (P<0.20 in univariate analysis; Table 2). After adjusting for these variables in a multivariate logistic regression model, patients with PSA were about 50% less likely to report a history of hayfever compared to control population and this difference almost reached statistical significance (adjusted OR=0.48 [CI 95% 0.23-1.03]; Table 3.). There was no difference in the likelihood of self-reported hayfever between PSA and PSO patients. (adjusted OR=0.91 [CI 95% 0.39-2.14])

Figure 1. Life-time prevalences of atopic disorders and sensitization in PSO, PSA and control groups



Abbreviations: PSO, psoriasis vulgaris; PSA, Psoriatic Arthritis, Increased total IgE, serum total IgE levels of more than 100 kU/L

P-values given were obtained using Chi-square test. On top of bars absolute numbers are given. Statistically significant results are printed in bold.

Asthma

The percentage of subjects reporting asthma was lowest in the PSA group followed by controls and PSO patients (3.6% vs. 9.5% vs. 9.8%; p= 0.031 and p=0.028 Figure 1). The difference between PSO and controls was not statistically significant. Potential confounder for this association was the age (Table 2) and after adjusting for it in a multivariate model, patients with PSA were approximately 70% less likely to have a history of asthma compared to controls. (adjusted OR=0.34 [CI 95% 0.13-0.92]; Table 3). The adjusted odds ratio for the comparison between PSA and PSO was 0.38 [CI 95% 0.14-1.04] suggesting a protective effect that was not statistically significant.

Of the 6 asthmatic PSA patients only one declared to have used inhalators in the past 12 months. In the control group 8 of 14 individuals with asthma reported having used inhalators. In the PSO group there were 2 inhalator users in 13 asthmatics.

Sensitization to common aeroallergens and total serum IgE

The proportions of PSO and control patients sensitized to common aeroallergens were similar (35.4 % vs. 34.0%), but significantly higher than the 19.7% detected in the group of PSA patients (p=0.003 and p=0.004, respectively). The difference between PSO and

Table 2. Uni-and multivariate logistic regression analysis of clinical and serological manifestations of atopy (n=448)

	History of clinical atopy				Serological atopy			
	Hayfever		Asthma		Sensitization to CAA		Increased IgE+	
	Crude OR (95%CI)	Adjusted OR*(95%CI)	Crude OR (95%CI)	Adjusted OR*(95%CI)	Crude OR (95%CI)	Adjusted OR*(95%CI)	Crude OR (95%CI)	Adjusted OR*(95%CI)
Sex(M)	0.72 (0.40-1.31)	N/A	1.45 (0.70-2.98)	N/A	**1.83 (1.20-2.79)**	**1.97 (1.27-3.07)**	1.49 (0.94-2.37)	**1.89 (1.11-3.21)**
Age	0.96 (0.94-0.98)	**0.96 (0.94-0.98)**	0.98 (0.96-1.00)	0.98 (0.96-1.01)	**0.97 (0.96-0.99)**	**0.97 (0.96-0.99)**	0.96 (0.97-1.00)	0.99 (0.97-1.01)
Methotrexate use	0.46 (0.19-1.11)	0.53 (0.20-1.42)	1.14 (0.50-2.61)	N/A	**0.56 (0.33-0.96)**	0.82 (0.42-1.60)	**0.49 (0.26-0.93)**	0.71 (0.33-1.56)
Biologic use	1.05 (0.39-2.8)	N/A	1.29 (0.43-3.87)	N/A	1.07 (0.54-2.11)	N/A	1.48 (0.73-3.01)	N/A
Smoking	0.87 (0.43-1.79)	N/A	0.79 (0.33-1.88)	N/A	1.00 (0.60-1.65)	N/A	1.65 (0.96-2.83)	1.27 (0.70-2.29)
PSO diagnosis	1.17 (0.62-2.21)	N/A	1.60 (0.77-3.32)	N/A	1.52 (0.98-2.36)	0.84 (0.49-1.42)	2.22 (1.38-3.56)	1.57 (0.83-2.96)
PSA diagnosis	0.57 (0.29-1.11)	0.78 (0.37-1.51)	**0.35 (0.14-0.86)**	**0.35 (0.14-0.89)**	**0.46 (0.30-0.73)**	**0.45 (0.24-0.83)**	**0.40 (0.23-0.67)**	0.59 (0.27-1.25)

Abbreviations: OR, Odds Ratio; 95%CI, 95% confidence intervals; PSO, psoriasis vulgaris; PSA, Psoriatic Arthritis, CAA, Common Aeroallergens, IgE, Immunoglubulin E, N/A; variables with an effect of p>0.20 in univariate analysis

**Adjusted for variables in left column.*

+Defined as total serum IgE > 100 kU/L

Statistically significant results are printed in bold.

controls was not statistically significant. Methotrexate use, sex and age were associated with sensitization to common allergens in univariate logistic regression analysis (p<0.20; Table 2). After adjusting for these variables the likelihood of sensitization was about less than half in PSA patients compared to controls (adjusted OR=0.43 [95%CI 0.23-0.83] Table 3). Similar results were found for PSA vs. PSO (adjusted OR=0.53 [95%CI 0.29-0.87]). Comparison of PSO versus control yielded a statistically non-significant difference (adjusted OR=1.10 [95%CI 0.60-2.02])

The proportions of subjects with total serum IgE levels > 100kU/L were 21.8%, 31.5% and 12.6% for controls, PSO and PSA groups respectively. The difference in the prevalence of increased IgE between PSA and controls was significant (p=0.028) which was also the case for comparison between PSA and PSO (p<0.001). Variables potentially confounding the associations with increased serum total IgE were sex, age, methotrexate use and smoking (Table 2). The adjusted OR for having increased total serum IgE in PSA vs. controls was 0.52 (95%CI 0.22-1.21; Table 3). Also, compared to PSO, PSA patients were significantly less likely to show elevated serum IgE levels (adjusted OR=0.35 [95%CI 0.17-0.72]).

Table 3. Uni-and multivariate binary logistic regression analyses comparing clinical and serological manifestations of atopy in PSO, PSA and control groups

Atopic features	**PSA vs PSO**		**PSA vs controls**		**PSO vs controls**	
	Crude OR (95%CI)	Adjusted OR* (95%CI)	Crude OR (95%CI)	Adjusted OR* (95%CI)	Crude OR (95%CI)	Adjusted OR* (95%CI)
Hayfever	0.61 (0.28-1.33)	0.91 (0.39-2.14)	0.53 (0.26-1.11)	0.48 (0.23-1.03)	0.87 (0.43-1.76)	0.80 (0.38-1.67)
Asthma	**0.34 (0.13-0.93)**	0.38 (0.14-1.04)	**0.35 (0.13-0.94)**	**0.34 (0.13-0.92)**	1.03 (0.47-2.28)	0.96 (0.43-2.15)
Sensitization to common aeroallergens	**0.45 (0.27-0.76)**	**0.53 (0.29-0.96)**	**0.48 (0.29-0.80)**	**0.43 (0.23-0.83)**	1.06 (0.65-1.74)	0.84 (0.49-1.45)
Total IgE >100ku/L	**0.31 (0.17-0.56)**	**0.35 (0.17-0.72)**	**0.50 (0.27-0.91)**	0.52 (0.22-1.21)	1.60 (0.93-2.72)	1.52 (0.79-2.93)

Abbreviations: PSO, psoriasis vulgaris; PSA, psoriatic arthritis; OR, odds ratio; 95%CI, 95% confidence intervals;

*multivariate logistic regression model was adjusted for age, sex, methotrexate use and current smoking where appropriate according to univariate logistic regression analysis from table 2. *Statistically significant results are printed in bold.*

Effect on clinical activity and course severity

To assess the impact of sensitization to common aeroallergens on clinical activity of arthritis in PSA the median scores of clinical PSA severity measures in sensitized and non-sensitized PSA patients were compared. The median DAS28 score in sensitized patients with PSA was nonsignificantly lower than in non-sensitized patients (2.12 (IQR 1.78-3.11) vs. 2.60 (IQR 1.98-3.35; p=0.190).

The median HAQ VAS score for pain in sensitized PSA patients was significantly lower than in non-sensitized patients (0.33 [IQR 0.21-1.02] vs 1.23 [IQR 0.56-1.86], p<0.001). Similar results were obtained for HAQ patient global VAS (19.0 [IQR 5.0-35.0] vs. 45.0 [IQR 19.0-64.0] p<0.001).

The median HAQ-DI score was significantly lower in sensitized patients with PSA (0.25 [IQR 0-0.75]) compared to the score in those non sensitized (0.75 [IQR 0.25-1.25], p=0.016). Further evaluation of these differences in a multivariate linear regression analysis revealed that the DAS28 score and HAQ-DI score were not significantly influenced by sensitization to common aeroallergens (Table 4). HAQ-VAS for pain and HAQ-VAS in PSA patients for patient global score were significantly influenced by sensitization to common aeroallergens (beta-coefficient -0.54 [95% CI -0.84 - -0.25] and -18.4 [95% CI -28.5 - -8.25] respectively.)

DISCUSSION

This study has consistently demonstrated a reduced prevalence of atopic features such as asthma (and to a lesser extent hayfever) and sensitization to common aeroallergens and increased total serum IgE in patients with PSA compared to PSO patients and controls. The antagonism between atopic disorders and autoinflammatory conditions has been found previously in type 1 diabetes mellitus[20-24] and in rheumatoid arthritis[9-11]. Our data suggest that immunological mechanisms in PSA may be more similar to those of rheumatoid arthritis than those of psoriasis vulgaris. The research on this subject in patients with psoriasis whether cutaneous or arthropathic has been limited to studies describing concurrent existence of atopic dermatitis or allergic contact dermatitis and cutaneous psoriasis[6, 25-27]. These studies have not shown antagonism on observational level between psoriasis and atopic dermatitis. So far, no studies have focused on patients with PSA in this matter. The two VAS HAQ scores assessing disease severity were significantly lower in sensitized PSA patients and this effect was greater than that of confounding variables included in the forward selection mode. The HAQ-DI score was also significantly lower in sensitized patients but this effect was diluted in linear multiple

Table 4. Linear uni- and multivariate regression analyses for clinical severity outcomes in 169 patients with psoriatic arthritis.

168 patients with psoriatic arthritis	DAS 28		HAQ-VAS for pain		HAQ-VAS global patient score		HAQ-DI	
	Univariate Beta-coefficient and 95% CI	Multivariate Beta-coefficient and 95% CI	Univariate Beta-coefficient and 95% CI	Multivariate Beta-coefficient and 95% CI	Univariate Beta-coefficient and 95% CI	Multivariate Beta-coefficient and 95% CI	Univariate Beta-coefficient and 95% CI	Multivariate Beta-coefficient and 95% CI
Sensitization to common aeroallergens	-0.27 (-0.70 - 0.16)	N/A	**-0.54 (-0.84 - -0.25)**	**-0.51 (-0.81 - -0.22)**	**-18.4 (-28.5 - -8.25**)	**-17.40 (-27.6 - -7.17**)	**-0.29 (-057 - -0.02)**	-0.16 (-0.48 – 0.17)
Age in years	0.01 (-0.01 - 0.02)	N/A	0.00 (-0.01 - 0.01)	N/A	-0.01 (-0.38 – 0.35)	N/A	**0.01 (0.00 - 0.02)**	0.01 (-0.10 – 0.01)
Sex (1= male,0=female)	**-0.53 (-0.85 - -0.20)**	**-0.51 (-0.83 - -0.18)**	-0.23 (-0.48 - 0.13)	-0.17 (-0.41 – 0.07)	**-7.40 (-15.9 - -1.02)**	-5.32 (-13.6 – 2.93)	**-0.30 (-0.52 - -0.08)**	-0.18 (-0.43 – 0.07)
Methotrexate use	**-0.26 (-0.59 - -0.08)**	-0.21 (-0.53 - 0.12)	0.04 (-0.21 - 0.29)	N/A	3.23 (-5.28 – 11.75)	N/A	0.08 (-0.14 – 0.30)	N/A
Duration of arthritis in years	0.01 (-0.01 - 0.03)	N/A	0.00 (-0.01 - -0.02)	N/A	0.06 (-0.45 – 0.58)	N/A	**0.02 (0.02 – 0.03)**	**0.02 (0 – 0.03)**
R^2 for multivariate analysis		0.07		0.10		0.10		0.06

Abbreviations: DAS28, Disease Activity Score for 28 joints; HAQ-VAS pain; Health Assessment Questionnaire Visual Analogue Scale pain score; HAQ-VAS patient global scale; idem dpatient global scale; HAQ-DI, idem Disability Index, R^2; coefficient of determination, 95% CI; 95% Confidence Intervals, N/A; variables with an effect of p>0.20 in univariate analysis

regression analysis. Disease activity score (DAS28) which is an arthritis symptom score did not differ between the atopic and non-atopic PSA patients. However, this study may have been underpowered to detect differences in disease severity

Immunological explanation

A mechanistic explanation for the found antagonism can not be deduced from our data. It is not clear on which level (e.g. genetic, cellular, proteomic or confounding) the cross regulation between atopy and psoriatic arthritis takes place. We can exclude simple genetics as a cause because the prevalences of family history of atopy in patients with psoriatic arthritis and controls were similar. Antagonism on cellular level, assuming an immune system skewed towards Th-2 responses in atopics, could provide a basic explanation. Previous studies have favoured this hypothesis. Nevertheless, unknown confounders can not be excluded. The difference in clinical and serological atopy between PSO and PSA is a striking finding which suggests a principal, rather than a gradual immunological difference between the two diseases.

Stengths and limitations

This is the largest observational study investigating the association between atopic features and PSO and the first to study frequency of atopy in PSA patients. The PSO and PSA patients have all been diagnosed by dermatologists and reumatologists, respectively, to minimize misclassification bias. Psoriasis is a clinical diagnosis easily made after physical examination and CASPAR criteria have a specificity and sensitivity of over 99% for PSA[28]. The Dutch areas where both recruiting hospitals are situated do not differ with respect to demographic characteristics and degree of urbanisation (Rotterdam vs Eindhoven). The equal percentages of family history of atopy in all groups suggest similarities in population composition for both hospitals. Therefore, the effect of the use of different hospitals departments for PSO and controls vs PSA on the study findings appears to be limited.

The selection of an appropriate control group is challenging and we choose patients with varicositas as controls because it is a common condition and not known to be associated with inflammatory diseases. There is no commonly accepted definition of the diagnosis of 'atopy'.[1] Since atopy is a clinical predisposition towards development of atopic dermatitis, atopic asthma and hayfever, we included self-report presence of hayfever and asthma based on validated questoinnaires, but excluded atopic dermatitis because it is more difficult to distinguish from other forms of dermatitis including psoriasis. In addition to clinical diagnosis, serum IgE directed to common inhalant allergens was added.[29] A limitation of all observational studies is establishing causality. However, it can be assumed that atopy preceded PSO and PSA because atopic disorders, in contrast

to PSO and PSA, occur during childhood.[30] It is therefore plausible that atopic disorders protect against the development of psoriatic arthritis and not vice versa, but, nevertheless, other longitudinal observational and fundamental studies need to confirm this hypothesis.

In this study, self-reported (history of) atopic dermatitis was excluded because of the clinical similarities to psoriasis, but it would be interesting to assess the life time prevalence of atopic dermatitis in psoriatics because skin is the target organ in both diseases

In conclusion, this is the first study reporting a reduced prevalence of atopic features in PSA and not PSO. Longitudinal cohort studies are needed to assess the causality of this relationship and fundamental research is needed to explore the underlying immunological mechanisms.

REFERENCES

1. Stern RS, Nijsten T, Feldman SR, Margolis DJ, Rolstad T. Psoriasis is common, carries a substantial burden even when not extensive, and is associated with widespread treatment dissatisfaction. J Investig Dermatol Symp Proc 2004;9:136-9.
2. Gelfand JM, Gladman DD, Mease PJ, et al. Epidemiology of psoriatic arthritis in the population of the United States. J Am Acad Dermatol 2005;53:573.
3. Ravindran V, Scott DL, Choy EH. A systematic review and meta-analysis of efficacy and toxicity of disease modifying anti-rheumatic drugs and biological agents for psoriatic arthritis. Ann Rheum Dis 2008;67:855-9.
4. Nestle FO, Kaplan DH, Barker J. Psoriasis. N Engl J Med 2009;361:496-509.
5. Steinman L. A brief history of T(H)17, the first major revision in the T(H)1/T(H)2 hypothesis of T cell-mediated tissue damage. Nat Med 2007;13:139-45.
6. Eyerich S, Onken AT, Weidinger S, et al. Mutual antagonism of T cells causing psoriasis and atopic eczema. N Engl J Med 2011;365:231-8.
7. Robinson DS. T-cell cytokines: what we have learned from human studies. Paediatr Respir Rev 2004;5 Suppl A:S53-8.
8. Rabin RL, Levinson AI. The nexus between atopic disease and autoimmunity: a review of the epidemiological and mechanistic literature. Clin Exp Immunol 2008;153:19-30.
9. Verhoef CM, van Roon JA, Vianen ME, Bruijnzeel-Koomen CA, Lafeber FP, Bijlsma JW. Mutual antagonism of rheumatoid arthritis and hay fever; a role for type 1/type 2 T cell balance. Ann Rheum Dis 1998;57:275-80.
10. Rudwaleit M, Andermann B, Alten R, et al. Atopic disorders in ankylosing spondylitis and rheumatoid arthritis. Ann Rheum Dis 2002;61:968-74.
11. Hilliquin P, Allanore Y, Coste J, Renoux M, Kahan A, Menkes CJ. Reduced incidence and prevalence of atopy in rheumatoid arthritis. Results of a case-control study. Rheumatology (Oxford) 2000;39:1020-6.

12. Rudwaleit M, Taylor WJ. Classification criteria for psoriatic arthritis and ankylosing spondylitis/axial spondyloarthritis. Best Pract Res Clin Rheumatol 2010;24:589-604.
13. Asher MI, Keil U, Anderson HR, et al. International Study of Asthma and Allergies in Childhood (ISAAC): rationale and methods. Eur Respir J 1995;8:483-91.
14. Burney PG, Luczynska C, Chinn S, Jarvis D. The European Community Respiratory Health Survey. Eur Respir J 1994;7:954-60.
15. Sanz ML, Prieto I, Garcia BE, Oehling A. Diagnostic reliability considerations of specific IgE determination. J Investig Allergol Clin Immunol 1996;6:152-61.
16. Wever AM, Wever-Hess J, van Schayck CP, van Weel C. Evaluation of the Phadiatop test in an epidemiological study. Allergy 1990;45:92-7.
17. Leung YY, Tam LS, Kun EW, Ho KW, Li EK. Comparison of 4 functional indexes in psoriatic arthritis with axial or peripheral disease subgroups using Rasch analyses. J Rheumatol 2008;35:1613-21.
18. Ujfalussy I, Koo E. Measurement of disease activity in psoriatic arthritis. Extended report. Z Rheumatol 2003;62:60-5.
19. Boulet LP, Turcotte H, Laprise C, et al. Comparative degree and type of sensitization to common indoor and outdoor allergens in subjects with allergic rhinitis and/or asthma. Clin Exp Allergy 1997;27:52-9.
20. Hermansson B, Holmgren G, Samuelson G. Juvenile diabetes mellitus and atopy. Hum Hered 1971;21:504-8.
21. Stromberg LG, Ludvigsson GJ, Bjorksten B. Atopic allergy and delayed hypersensitivity in children with diabetes. J Allergy Clin Immunol 1995;96:188-92.
22. Decreased prevalence of atopic diseases in children with diabetes. The EURODIAB Substudy 2 Study Group. J Pediatr 2000;137:470-4.
23. Douek IF, Leech NJ, Bingley PJ, Gale EA. Eczema and Type 1 diabetes. Diabet Med 2002;19:174-5.
24. Cardwell CR, Shields MD, Carson DJ, Patterson CC. A meta-analysis of the association between childhood type 1 diabetes and atopic disease. Diabetes Care 2003;26:2568-74.
25. Pigatto PD. Atopy and contact sensitization in psoriasis. Acta Derm Venereol Suppl (Stockh) 2000:19-20.
26. Clark AR, Sherertz EF. The incidence of allergic contact dermatitis in patients with psoriasis vulgaris. Am J Contact Dermat 1998;9:96-9.
27. Rocken M, Link C, Breit R. [The incidence of atopic symptoms in patients with psoriasis] Haufigkeit atopischer Symptome bei Patienten mit Psoriasis. Hautarzt 1991;42:684-6.
28. Chandran V, Schentag CT, Gladman DD. Sensitivity and specificity of the CASPAR criteria for psoriatic arthritis in a family medicine clinic setting. J Rheumatol 2008;35:2069-70; author reply 70.
29. Frith J, Fleming L, Bossley C, Ullmann N, Bush A. The complexities of defining atopy in severe childhood asthma. Clin Exp Allergy 2011;41:948-53.
30. Spergel JM, Paller AS. Atopic dermatitis and the atopic march. J Allergy Clin Immunol 2003;112:S118-27.
31. Sporik R, Holgate ST, Platts-Mills TA, Cogswell JJ. Exposure to house-dust mite allergen (Der p I) and the development of asthma in childhood. A prospective study. N Engl J Med 1990;323:502-7.

Chapter 2.2

Systemic B-cell abnormalities in patients with atopic dermatitis

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To the Editor:

With great interest we read the article of Czarnowicki *et al.*, in which local and systemic B-cell abnormalities in patients with atopic dermatitis (AD) were presented.[1] Specifically, relative expansions of transitional, CD27+IgD+ memory and IgE-expressing B cells were found. We recently proposed a reproducible approach for identifying IgE-expressing B cells, and showed that the CD27-IgE+ memory B-cell subset was expanded with increased levels of somatic hypermutations in patients with AD, while CD27+IgE+ memory B-cell and plasma cell frequencies were normal.[2] In line with a multitude of other chronic immune diseases, Czarnowicki *et al.* observed more general B-cell abnormalities in patients with AD.[1]

Despite the extensive analysis, no attention was paid to the non-Gaussian distribution of cell frequencies,[2-4] and no absolute numbers were reported. Such analyses as well as reproducibility in independent cohorts are necessary for proper understanding of disease pathology and for patient stratification to improve therapy success.

Therefore, we here analyzed the blood B-cell compartment of our previously published cohorts of patients with AD and patients with psoriasis.[2] Within total CD19+ B cells, the naive, memory and plasma blast cell subsets were defined by multi-color flow cytometry (Figure 1).[2, 5] In addition, IgM-only and Ig-class switched memory B cells were defined within the IgD- events. This allowed for reliably detection of IgE expressing B-cells without contamination of IgE-binding B-cells via the low FcεRII (CD23). No differences were found between healthy controls and psoriasis patients. AD patients did show significantly lower frequencies of CD27+IgD+ and CD27+IgD-IgM+ 'IgM-only' memory B cells, and an increase in CD27-IgE+ memory B cells (Figure 1C).[2]

Upon calculation to absolute cell numbers per microliter of blood, only the decrease of IgM-only memory B cells remained significant for AD patients (Figure 1D); no difference was seen for CD27+IgD+ B cells and the CD27-IgE+ population was not significantly increased (p=0.08).

Thus, in contrast to Czarnowicki et al. we did not find expansions of transitional B cells, CD27+IgD+ memory B cells and total or IgE+ plasma blasts in AD patients. Our cohort size was slightly smaller than that of Czarnowicki et al. (23 vs 34 subjects), but with a similarly large age range (16-79yr vs 18-74yr) and an almost equal Female/Male ratio.[1, 2] While 15/23 patients had elevated serum IgE levels, these did not range as high as in the Czarnowicki cohort, which might account in part for the difference between our results.

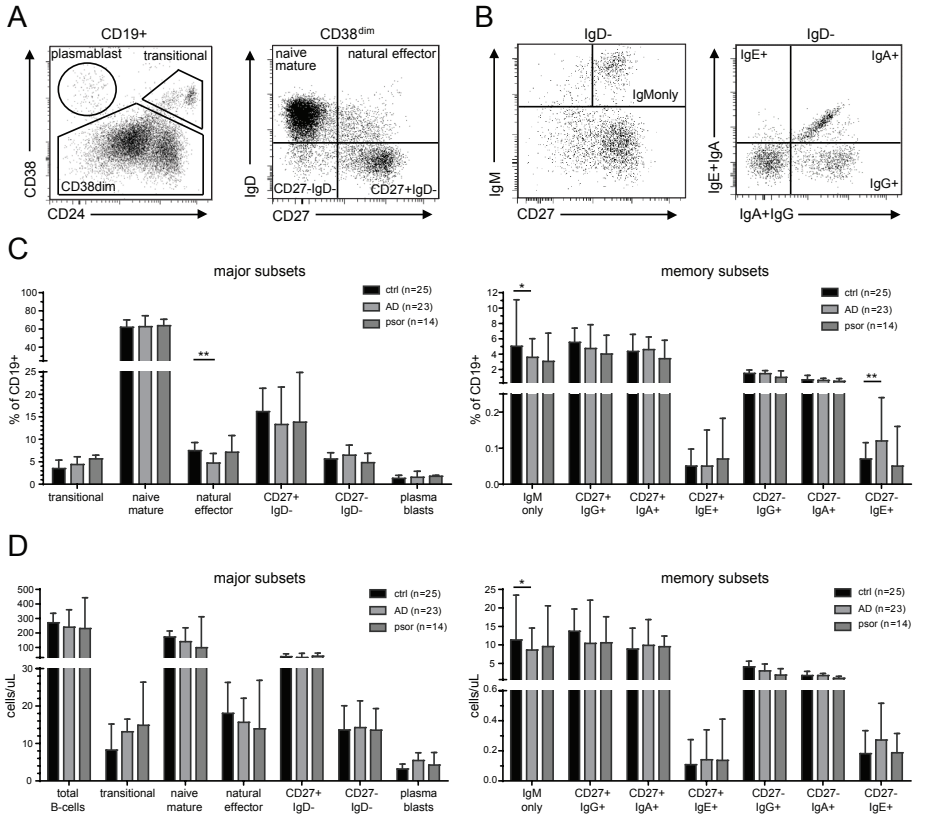

Figure 1. Absolute numbers and relative distributions of B-cell subsets in healthy adults, patients with atopic dermatitis and patients with psoriasis. A. Definition of the major naive and memory B-cell subsets and plasma cells with flow cytometric immunophenotyping. B. Further subsetting of IgD- memory B cells based on differential expression of CD27 and the remaining 4 Ig isotypes.[2, 5] C. Relative distributions of B-cell subsets. D. Absolute numbers of total B cells and B-cell subsets. *, p<0.05; **, p<0.01; Mann-Whitney U test.

Another explanation could lie in the fact that we here depicted the medians and calculated statistics with the non-parametric Mann-Whitney U test, rather than mean values and Student's t-test. We advocate a standardized analysis procedure to take into account non-Gaussian distributions, as well as calculation of absolute cell numbers to prevent misinterpretation of changes in co-dependent variables.[6, 7] Furthermore, interpretation of B-cell immunophenotyping can be improved by consensus nomenclature of the B-cell compartment.[5, 8]

In conclusion, the different observations from the Czarnowicki cohort and ours warrant further detailed analysis of B-cell phenotypes in AD patients, and especially whether these correlate with specific clinical findings. These detailed studies would be necessary

to establish whether AD patients have general abnormalities in their B-cell compartment or if these are mostly limited to the expansion of IgE-expressing B cells, as this could have major implications for future therapeutic approaches.

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REFERENCES

1. Czarnowicki T, Gonzalez J, Bonifacio KM, Shemer A, Xiangyu P, Kunjravia N, et al. Diverse activation and differentiation of multiple B-cell subsets in patients with atopic dermatitis but not in patients with psoriasis. J Allergy Clin Immunol 2015.
2. Berkowska MA, Heeringa JJ, Hajdarbegovic E, van der Burg M, Thio HB, van Hagen PM, et al. Human IgE(+) B cells are derived from T cell-dependent and T cell-independent pathways. J Allergy Clin Immunol 2014; 134:688-97 e6.
3. Jansen MA, van den Heuvel D, van Zelm MC, Jaddoe VW, Hofman A, de Jongste JC, et al. Decreased memory B cells and increased CD8 memory T cells in blood of breastfed children: the generation R study. PLoS One 2015; 10:e0126019.
4. van den Heuvel D, Driessen GJ, Berkowska MA, van der Burg M, Langerak AW, Zhao D, et al. Persistent subclinical immune defects in HIV-1-infected children treated with antiretroviral therapy. AIDS 2015; 29:1745-56.
5. Berkowska MA, Driessen GJ, Bikos V, Grosserichter-Wagener C, Stamatopoulos K, Cerutti A, et al. Human memory B cells originate from three distinct germinal center-dependent and -independent maturation pathways. Blood 2011; 118:2150-8.
6. Maecker HT, McCoy JP, Nussenblatt R. Standardizing immunophenotyping for the Human Immunology Project. Nat Rev Immunol 2012; 12:191-200.
7. van den Heuvel D, Jansen MA, Dik WA, Bouallouch-Charif H, Zhao D, van Kester KA, et al. Cytomegalovirus- and Epstein-Barr Virus-Induced T-Cell Expansions in Young Children Do Not Impair Naive T-cell Populations or Vaccination Responses: The Generation R Study. J Infect Dis 2015.
8. Kaminski DA, Wei C, Qian Y, Rosenberg AF, Sanz I. Advances in human B cell phenotypic profiling. Frontiers in Immunology 2012; 3.

Chapter 3

Atopy and cancer

Chapter 3.1

Non-melanoma skin cancer: the hygiene hypothesis

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ABSTRACT

Protection against ultraviolet radiation-induced DNA-damage in the skin is not only provided by the pigmentary system. The epidermal barrier consisting of stratum corneum keratinocytes, filaggrin and other proteins is an additional component of the UV-shield. Disruption of the epidermal barrier through frequent body cleansing with soaps and cosmetics may increase the risk of non-melanoma skin cancer.

INTRODUCTION

Since the 1950s it is known that excessive exposure to ultraviolet radiation (UVR) is associated with an increased risk of nonmelanoma skin cancer [1]. Non-melanoma skin cancer is the term which covers basal cell carcinoma (BCC) and squamous cell carcinoma (SCC). UVB and UVA are both oncogenic and immunosuppressant, but UVB mostly acts through direct mutagenic oncogenesis while UVA's effects mainly impair the immunosurveillance. The current leading theory, fueled by overwhelming epidemiologic evidence is that epidermal protection against skin cancers is solely provided by the pigmentary system. This barrier of melanin produced by melanocytes in the basal layer of the skin is transferred to the keratinocytes via melanosomes. The mismatch between skin phototype (degree of pigmentation) and UVR exposure has become a truism to explain the rise in incidence of non-melanoma skin cancer. This mismatch is the single most important risk factor for non-melanoma skin cancer which has left little room for research on alternative risk factors. The currently failing prevention programs necessitate the analysis of preventable causes [2]. Here, we propose epidermal barrier dysfunction through personal hygiene habits as a new risk factor for the development of non-melanoma skin cancers.

Why pigmentation only does not suffice

Melanin is thought to protect the skin from the DNA-damaging effects of UV-light. The lower prevalence of non-melanoma skin cancer in dark skinned individuals underscores this. Nevertheless, the existence of DNA-repair mechanisms in the skin indicate that pigment only may not suffice to protect us. The rare cases of African patients with xeroderma pigmentosum illustrate this in vivo. These patients develop squamous cell carcinomas in the typical photodistributed locations very early in their lives [3].While a darker skin type in healthy Africans does offer protection against nonmelanoma skin cancer, facultative pigmentation as seen in people of European ancestry offers no protection against development of UV-induced pyrimidine dimers [4]. Pheomelanin even induces apoptosis of keratinocytes as it acts as a photosensitizer after exposure to UVB [5]. Although the pigmentary system protects us from acute sunburn, the development of non-melanoma skin cancer does not threaten the survival of individuals before reproductive age. In the anogenital area, the squamous cell cancer arises without significant, previous UVB-exposure. This is explained by the oncogenic effects of human papillomavirus and indicates the existence of alternative and complementary routes of pathogenesis from which melanin offers no protection.

Stratum corneum as UVR-barrier

There is more in the epidermis than pigment only to block the UVR. The most outer layer of the skin, stratum corneum, is the first component of the skin to absorb the photons. It is known that low doses of UVR from the ambient spectrum may not even penetrate stratum corneum [6]. Thickening of the stratum corneum is the most important adaptation to acute UVR-exposure in humans [7]. In a study with vitiligo patients, the skin stripped of stratum corneum was significantly more sensitive to UVB in terms of mean erythema dose than non-stripped skin [8]. Besides the dead keratinocytes and their contents the photons from UVR also encounter a barrier of many proteins, amino acids, ceramides and lipids in stratum corneum. Stratum corneum, together with all these components is also referred to as the epidermal barrier. This barrier is held in place by a scaffold of filaggrin. Filaggrin is a protein which contributes to compaction of stratum corneum and cohesion between its constituents [9]. Impairments in the functioning of filaggrin, its precursors or derivatives have been linked to inflammatory skin disorders like atopic dermatitis but may also lead to a disrupted epidermal UVR-barrier. A study with a human skin model showed increased UVB-induced apoptosis after

knockdown of filaggrin expression [10]. Conversely, in response to UVB-exposure filaggrin expression is upregulated together with other components of the epidermal barrier [11]. Derivatives of the filaggrin metabolism also may provide protection against UVB induced effects. Filaggrin cleavage by caspase-14 is required for its proper functioning. A major derivative of the cleavage process of filaggrin by caspase-14 is urocanic acid. Urocanic acid provides protection against dehydration and low-level protection against UVB-induced DNA-damage [12]. The skin of caspase-14-deficient mice is highly sensitive to the formation of pyrimidine dimers after exposure to UVB compared to wild type mice [13]. These data strongly suggest that the stratum corneum provides an additional component for the total UVR barrier.

EPIDERMAL BARRIER DYSFUNCTION AND SKIN CANCER

Previously mentioned studies have demonstrated increased sensitivity to UVR in dysfunctional stratum corneum but there is also circumstantial evidence that defects in the stratum corneum may actually lead to non-melanoma skin cancer. Patients with skin disorders primarily related to epidermal barrier dysfunction have been of particular interest. In a Danish cohort of 31,330 patients with atopic dermatitis, a common disorder of the epidermal barrier the standardized incidence ratios for BCC and SCC were 1.41 [95% CI 1.07-1.83] and 2.48 [95% CI 1.00-5.11] respectively [14]. Patients with epidermal barrier dysfunction are also frequently sensitized to various allergens because of the

more permeable epidermis. They develop antibodies of the IgE-type. Patients who have multiple squamous cell carcinomas have these antibodies more often (odds ratio 3.82 [95% CI 1.05-13.88]) [15]. This suggests that patients with an impaired epidermal barrier, which is indicated by the presence of sensitization to common allergens, are at higher risk of developing non-melanoma skin cancer. Same results can be found in non-vulgaris ichthyosis and Netherton's syndrome. These rare disorders are extreme examples of epidermal barrier dysfunction. Patients with these disorders show an increased susceptibility to squamous cell carcinoma [16]. In Netherton's syndrome a mutation in SPINK5 leads to impaired function of a serine protease inhibitor. The proteases which it inhibits are involved in desquamation and remodeling of the stratum corneum and processing of filaggrin. These patients often have an atopic dermatitis-like phenotype and exhibit high serum-levels of IgE.

How the hygiene may contribute

The epidermis is continuously exposed to physical and chemical assaults. Of all physical agents application of detergent is the most damaging to the anatomy and function of stratum corneum [17]. Through personal hygiene habits our skin is frequently exposed to warm water during showering and to detergents from soaps and cosmetics. This hygiene behaviour and lifestyle is a relatively recent development in human evolution and our skin may not be adapted to it. Dermatologists typically discourage vigorous use of soap and frequent showering for patients with atopic dermatitis. From anecdotal evidence we know that this may worsen the disease. However, no high-level evidence exists to back this practice up. Even less is known about the influence of hygiene and cosmetic products on the skin in healthy individuals. The use of showering gels impairs the function and damages the stratum corneum [18]. This is probably mostly attributable to the effects of sodium lauryl sulphate which is ubiquitous in commercially available products [19]. When in contact with skin it initiates denaturation of the epidermal proteins which subsequently leads to an impaired epidermal barrier function [20]. Filaggrin is water-soluble. Therefore, even exposure to only water during washing may rinse of significant amounts of the epidermal scaffolding which takes some time to replace. We postulate that frequent exposure to showering and detergents in soaps and cosmetics, as habitually used in modern times, lowers epidermal UVR-blocking capacities and increases the risk of non-melanoma skin cancer.

TESTING THE HYPOTHESIS

Future studies may encounter some problems. It will be difficult for the fundamental scientist to define hygienic behaviour in animal models and dysfunctional skin barrier.

This makes the results hard to interpret and to extrapolate to humans and clinically relevant endpoints. Genetic studies may reveal polymorphisms influencing the epidermal barrier and the development of nonmelanoma skin cancer but the practice of hygiene is obviously a cultural phenomenon which cannot be taken into account within these studies. Functional testing with measuring of the epidermal barrier function is not feasible in large groups of patients and the time until the development of skin cancer may blur the results. Also, the amount of damage to the skin barrier through external factors needs defining especially in longitudinal cohort studies. The testing of our hypothesis therefore necessitates research involving different areas. In vitro studies should focus on how stratum corneum and filaggrin protect us against UVR-induced DNA-damage and how this may be impaired by environmental factors. Genetic studies are needed to help form new hypotheses. Epidemiologic studies may show or dispute associations between diseases of the epidermal barrier such as atopic dermatitis and non-melanoma skin cancer. This is a cumbersome field because of many potential confounders such as use of immunosuppressant medication and treatment with UVR. The role of hygiene and cosmetics as well as exposure to yet unknown environmental factors should be explored. After the identification of relevant epidemiologic, genetic and biologic parameters interventional studies can be initiated. Though randomizing patients into "hygienic" and "less hygienic" groups may be unthinkable for longer periods of time, it is feasible to have two groups of individuals practicing different hygiene behaviours during holidays when disproportionate exposure to UVR takes place. Finally, the gained knowledge can be implemented through prevention studies.

REFERENCES

1. Blum HF. Sunlight as a casual factor in cancer of the skin of man. J Natl Cancer Inst 1948;9:247-58.
2. Madan V, Lear JT, Szeimies RM. Non-melanoma skin cancer. Lancet 2010;375:673-85.
3. Chidzonga MM, Mahomva L, Makunike-Mutasa R, Masanganise R. Xeroderma pigmentosum: a retrospective case series in Zimbabwe. J Oral Maxillofac Surg 2009;67:22-31.
4. Niggli HJ. Comparative studies on the correlation between pyrimidine dimer formation and tyrosinase activity in cloudman S91 melanoma cells after ultraviolet-irradiation. Photochem Photobiol 1990;52:519-24.
5. Takeuchi S, Zhang W, Wakamatsu K, et al. Melanin acts as a potent UVB photosensitizer to cause an atypical mode of cell death in murine skin. Proc Natl Acad Sci USA 2004;101:15076-81.
6. Phan TA, Halliday GM, Barnetson RS, Damian DL. Melanin differentially protects from the initiation and progression of threshold UV-induced erythema depending on UV waveband. Photodermatol Photoimmunol Photomed 2006;22:174-80.
7. Pearse AD, Gaskell SA, Marks R. Epidermal changes in human skin following irradiation with either UVB or UVA. J Invest Dermatol 1987;88:83-7.

8. El-Khateeb EA, Ragab NF, Mohamed SA. Epidermal photoprotection: comparative study of narrowband ultraviolet B minimal erythema doses with and without stratum corneum stripping in normal and vitiligo skin. Clin Exp Dermatol 2011;36:393-8.
9. Sandilands A, Sutherland C, Irvine AD, McLean WH. Filaggrin in the frontline: role in skin barrier function and disease. J Cell Sci 2009;122:1285-94.
10. Mildner M, Jin J, Eckhart L, et al. Knockdown of filaggrin impairs diffusion barrier function and increases UV sensitivity in a human skin model. J Invest Dermatol 2010;130:2286-94.
11. Hong SP, Kim MJ, Jung MY, et al. Biopositive effects of low-dose UVB on epidermis: coordinate upregulation of antimicrobial peptides and permeability barrier reinforcement. J Invest Dermatol 2008;128:2880-7.
12. Barresi C, Stremnitzer C, Mlitz V, et al. Increased sensitivity of histidinemic mice to UVB radiation suggests a crucial role of endogenous urocanic acid in photoprotection. J Invest Dermatol 2011;131:188–94.
13. Denecker G, Hoste E, Gilbert B, et al. Caspase-14 protects against epidermal UVB photodamage and water loss. Nat Cell Biol 2007;9:666-74.
14. Jensen AO, Svaerke C, Kormendine Farkas D, Olesen AB, Kragballe K, Sorensen HT. Atopic dermatitis and risk of skin cancer: a Danish nationwide cohort study (1977-2006). Am J Clin Dermatol 2012;13:29-36.
15. Wiemels JL, Wiencke JK, Li Z, Ramos C, Nelson HH, Karagas MR. Risk of squamous cell carcinoma of the skin in relation to IgE: a nested case-control study. Cancer Epidemiol Biomarkers Prev 2011;20:2377-83.
16. Natsuga K, Akiyama M, Shimizu H. Malignant skin tumours in patients with inherited ichthyosis. Br J Dermatol 2011;165:263-8.
17. Tagami H, Kobayashi H, Zhen XS, Kikuchi K. Environmental effects on the functions of the stratum corneum. J Invest Dermatol Symp Proc 2001;6:87-94.
18. Gloor M, Wasik B, Gehring W, Grieshaber R, Kleesz P, Fluhr JW. Cleansing, dehydrating, barrier-damaging and irritating hyperaemising effect of four detergent brands: comparative studies using standardised washing models. Skin Res Technol 2004;10:1-9.
19. Fluhr JW, Kelterer D, Fuchs S, et al. Additive impairment of the barrier function and irritation by biogenic amines and sodium lauryl sulphate: a controlled in vivo tandem irritation study. Skin Pharmacol Physiol 2005;18:88-97.
20. Wolf R, Parish LC. Effect of soaps and detergents on epidermal barrier function. Clin Dermatol 2012;30:297-300.

Chapter 3.2

Atopic dermatitis is not associated with actinic keratosis: cross-sectional results from the Rotterdam study

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ABSTRACT

Background

Epidermal barrier impairment and altered immune system in atopic dermatitis (AD) may predispose to ultraviolet induced DNA damage.

Objective

To study the association between atopic dermatitis and actinic keratosis (AK) in a population-based cross-sectional study.

Methods

AD was defined by modified criteria of the U.K. working party's diagnostic criteria. The AKs were diagnosed by physicians during a full-body skin examination and keratinocyte cancers were identified via linking to the national pathology database. The results were analysed in adjusted multivariable and multinomial models.

Results

Lower proportion of subjects with AD had AKs than without AD (16% vs 24%, p=0.002; unadjusted OR 0.60; 95% CI, 0.42-0.83, adjusted 0.74; 95% CI, 0.51-1.05, fully adjusted 0.69; 95 CI, (0.47-1.07)). In a multinomial model AD patients were less likely to have 10 or more AKs (adjusted OR 0.28; 95% CI, 0.09-0.90). No effect of AD on BCC and SCC was found (adjusted OR 0.71; 95% CI, 0.41-1.24 and adjusted OR 1.54; 95% CI, 0.66-3.62).

Conclusion

AD in community dwelling patients is not associated with AK.

What is already known

* Patients with severe AD treated with UV and immunosuppressants are at an increased risk of keratinocyte malignancies.

What does this study add?

* Community dwelling, average AD patients do not have more AKs or KCs.
* There is no support for extra AK and KC screening in patients with mild to moderate AD.

Abbreviations

AD; Atopic dermatitis
AK; Actinic keratosis
AR; Allergic rhinitis
BCC; Basal cell carcinoma
CI; Confidence interval
IRR; Incidence rate ratio
KC; Keratinocyte cancer
OR; Odds ratio
RS; Rotterdam study
SCC; Squamous cell carcinoma
UVB; Ultraviolet-B
UVR; Ultraviolet radiation
95% CI; 95% confidence interval

INTRODUCTION

Atopic dermatitis (AD) is one of the most prevalent inflammatory skin conditions affecting up to 30% of infants and up to 10% of adults and its incidence has tripled in the past three decades.[1] Atopic dermatitis results from an impaired epidermal barrier, mostly due to mutations in the filaggrin gene, combined with an overactive, Th2-polarized immune system. [2] It has been shown that the defective barrier may predispose AD patients to more ultraviolet radiation (UVR) induced DNA damage and subsequently increase the risk of keratinocyte cancers (KCs). There is, however, also ample evidence that increased epithelial turn-over and enhanced immunosurveillance actually protect against carcinogenesis in the atopic skin.[6,7]

KCs are common and their incidence is still increasing, which is most likely due to changes in UV exposure patterns. [5] Basal cell carcinoma (BCC) is the most common cancer in people of European ancestry. Squamous cell carcinoma (SCC) occurs at a comparable rate with melanoma, especially in elderly individuals and those chronically exposed to the sun. [8, 9] Actinic keratosis (AK) is a keratinocyte premalignancy and is also highly prevalent (25% of Dutch inhabitants older than 50 years of age) and is associated with high levels of cumulative UV exposure. [10,11]

In several mice studies, altered filaggrin expression and metabolism have been demonstrated to lead to an increased susceptibility to UVB-induced DNA damage.[4,5,12-14] In humans the results are inconsistent and the studies lack clear definition of exposure. Filaggrin loss-of-function mutations were not associated with incident skin cancer in a Danish cohort of 13376 genotyped individuals[15]. Three retrospective registry based cohort studies observed a 1.5 to 2.5 folds increased risk of developing KC in patients with AD, but these studies suffered from substantial residual confounding.[16-18] In contrast, four small retrospective studies suggested no association between AD and KC[19-22] and one mail survey including 254 AD patients suggested that AD had a protective effect on the development of KC.[23] None of these observational studies reported the association between AD and AK. The objective of this study was to investigate whether subjects with AD have more AKs than non-AD individuals in a population-based Dutch cohort (i.e. the Rotterdam Study). [24]

METHODS

Study design and study population

The Rotterdam study (RS) is an ongoing prospective, Dutch population-based cohort study that follows the inhabitants of the Ommoord district of Rotterdam, the Netherlands since 1990. [24]

Currently there are 14926 participants in the RS. The RS reflects the population of the Netherlands in terms of co-morbidities and health consumption. Dermatologic screening was introduced in August 2010. Since then, FBSE (full body skin examination), with the exception of the feet and the skin covered by socks and underwear, respectively are being conducted by four dermatologists trained physicians focussing on the most common skin diseases such as skin (pre-)malignancies, atopic dermatitis, hand eczema, psoriasis, and varicose veins. These were a predefined set of findings and diagnoses. All participants of the RS are to undergo FBSE consecutively without a particular order. Participants who had undergone FBSE were included in the present study. All were above 50 years of age and 95% were Caucasian.

The RS is approved by the Medical Ethics Committee of the Erasmus University Medical Centre and The Netherlands Ministry of Health. All patients participating in the study gave written informed consent. This study is a cross-sectional study drawn from the Rotterdam study.

Exposure: Atopic dermatitis

The participants were interviewed at home and FBSE was performed by trained physicians in a medical research facility in the centre of the district. No validated diagnostic criteria exist for adults with AD. However, in our practice, adults are diagnosed with AD on a daily basis. Therefore the commonly used UK working party's criteria, designed for a paediatric population needed adjusting before they could be applied to our elderly population. [25] These guidelines comprise one major criterion plus three or more minor criteria. 'An itchy skin condition' is the must have criterion and the minor criteria are: 'history of involvement of the skin creases', 'a personal history of asthma or hay fever', 'a history of generally dry skin in the last year', 'visible flexural eczema' and 'onset under 2 years of age.

Because of the possible confusion of sun damaged skin and hyperkeratosis of AK with xerosis and because of the high prevalence of dry skin in the elderly (56% in patients in the RS), the item of 'history of generally dry skin' was excluded from our AD definition. [26] The personal history of other atopic diseases was extended with 'house dust

mite allergy', because it is strongly related to atopic diseases.[27] Furthermore sites of visible eczema were adapted. In adults, atopic dermatitis also occurs on the hands and the trunk.[28] We therefore included these localisations. Finally the onset of age was prolonged to twelve years of age, because most patients develop AD before this age.[25] Atopic dermatitis could contribute to the development of AK's through either the defect in the epidermal barrier or the chronic inflammation in the skin. In order to be able to differentiate between these two hypotheses we performed analyses on AD criteria separately. In the case of the barrier-model one would expect all patients with AD to have an increased risk while in the case of chronic inflammation-model patients with current, i.e. long standing AD would be at more risk.

Outcome: Actinic keratosis

As previously reported in a prevalence study within RS, AKs were diagnosed by trained physicians and were defined as a rough (keratotic) lesion with adherent scaling and erythema, not fitting other diagnoses.[11] The cumulative number of observed AKs was divided into 4 categories: 0, 1-3, 4-9 or ≥10 AKs. Limiting the reporting of AK to categories instead of a continuous variable prevents inter-rater variability to some degree and makes for a clinically more relevant outcome corresponding with: no AK, mild, moderate and severe AKs.

Outcome: keratinocyte cancer

As described previously, the BCCs and SCCs were histologically confirmed by linking the RS participants to PALGA, a network of histo- and cytopathology which administrates the information of all biopsies and excisions from 1991 onward on a national level.[11] If at the time of the skin examination, suspicious lesions were observed they were biopsied, treated and included if pathologically confirmed as BCC or SCC.

Covariates

Previously identified risk factors for AK such as gender, age and pigmentation status were integrated in our analyses.[11] Pigmentation status was defined as a combination of three variables: hair colour at young age, eye colour and skin colour. The variable 'hair colour' contained 4 categories (fair/blonde, dark blonde/brown, red and black), 'eye colour' 3 categories (blue, intermediate, brown) and 'skin colour' had 6 options (albino, white, white to olive, light brown, brown, dark brown to black). The sum of the variables was recoded into three categories of light, medium or dark pigmentation status. Male baldness at the time of the skin examination was also recorded as a binary variable. Other possible risk factors for AKs were available from interview data: living in a sunny country for >1 year, the use of immunosuppressive medication , smoking, wearing sunglasses

and hats for protection, and the number of life time sunburns. The first three questions had binary responses and the last two were categorized as never/almost never, often/not always and always, and the absolute number of sunburns. These variables are not known for an association with AD, but because of their importance as a traditional risk factor for AKs, they were taken into account and they were used in the fully adjusted model.[27] Finally, we only included possible confounders with effects greater than 10% on the association between atopic dermatitis and AK in the main adjusted model (i.e. bivariate approach for selection of confounders).[29]

Statistical analysis

The null hypothesis was that AD is not associated with AK in a population based sample. The sample size for this study was calculated a priori and was based on a power of 80%, an altered risk of one third (OR of ≤0.66 or ≥1.33), based on an estimated AD prevalence of 10%. The calculated sample size (number of cases) for these ORs lies between 829 and 1253. It was powered for a univariate analysis for the association between AK and AD as SCC and BCC are not prevalent enough to provide over 829 cases. The primary analyses were performed by univariable and multivariable binary logistic regression looking at the association between AD and AK, SCC and BCC, separately. In a multinomial logistic regression model, the number of AKs was also analysed by categories namely 1-3 AKs, 4-9 AKs and >10 AKs, to assess whether the number of AKs changed the effect in relation to AD. Sensitivity analysis was performed to investigate whether having active dermatitis or past history of it or history of atopy was driving a potential association and whether altering the definition of exposure would alter the OR of the association. Individuals with missing variables relevant for the analyses were excluded. Values of $p<0.05$ were considered significant. All data were analysed using IBM SPSS 21.0, except for the sample size calculation; PS Power and Sample Size Calculations Version 3.0.

RESULTS

Patient characteristics

An FBSE and home interview were carried out in a total of 4,375 participants. The proportion of females in the cohort was 56% and the mean age was 67.6 years (range 53-100) (Table 1). Of the 4,375 participants, 24% had one or more AK of which 56.9% had 1-3 AKs, 22.8% had 4-9 and 20.4% had more than 10 AKs. The mean age of subjects with AKs was significantly higher than those without AKs, 73 vs 65.9 years ($p<0.01$).

Table 1. Frequencies and characteristics of the study population*

	Cases/AK (N=1050)		Controls/No AK (N=3325)		P-value
Characteristic	**N**	**(%)**	**N**	**%**	
AD criteria					P = 0.002
No	1006	(95.8)	3107	(93.4)	
Yes	42	(4.0)	218	(6.6)	
Missing	2	(0.2)	0	(0.0)	
Gender					P < 0.001
Women	444	(42.3)	1988	(59.8)	
Men	606	(57.7)	1337	(40.2)	
Age					P < 0.001
Mean age in years (SD)	73.0		65.9		
<70	379	(36.1)	2373	(71.4.)	
70-79,99	483	(46.0)	764	(23.0)	
≥80	188	(17.9)	188	(5.7)	
Pigmentation status (eye, hair and skin colour)					P < 0.001
Dark	55	(5.2)	450	(13.5)	
Medium	667	(63.5)	2190	(65.9)	
Light	260	(24.8)	528	(15.9)	
Data missing	68	(6.5)	157	(4.7)	
Tendency to develop sunburn					P < 0.001
Low	521	(49.6)	2165	(65.2)	
High	366	(34.9)	947	(28.5)	
Data missing	163	(15.5)	210	(6.3)	
History of living in a sunny country >1 year					P = 0.003
No	820	(78.1)	2783	(83.7)	
Yes	73	(7.0)	367	(11.0)	
Data missing	157	(15.0)	175	(5.3)	
Sun-protective behaviour					P < 0.001
Never/almost never	255	(24.3)	1151	(34.6)	
Often/not always	298	(28.4)	937	(28.2)	
Always	340	(32.4)	1062	(31.9)	
Data missing	157	(15.0)	175	(5.3)	

* cumulative percentages may not be 100% in all cases because of rounding

AD and AK

Of 4375 screened participants 6.3% met the AD-criteria. A significantly lower proportion of people with atopic dermatitis had actinic keratosis compared to those without atopic dermatitis (16% vs 24%, p=0.002; unadjusted 0.60; 95% CI, 0.42-0.83) (Table 2). Of all the

available potential confounders (Table 2), only age and gender changed the OR between AD and AK by at least 10% and were, therefore, included in the main multivariate model. After adjusting for age and gender, the association between AD and AK was not significant (adjusted OR 0.74; 95% CI, 0.51-1.05; Table 2). Comparable OR was found in the fully adjusted model 0.69; 95% CI, (0.47-1.07). In the multinomial model taking the AK categories into account, 1-3 AKs and 4-9 AKs showed no significant association with AD in both the unadjusted and age and gender adjusted and fully adjusted analyses (Table 2). In contrast to these low-number AK categories, AD patients were approximately 78% less likely to have 10 or more AKs compared to those without AD (adjusted OR 0.28; 95% CI 0.09-0.90). However this association did not remain significant in the fully adjusted model. The decreasing OR followed a statistically significant trend (p<0.001) (table2).

Table 2. Binary logistic regression of the association between AD and AK, unadjusted and adjusted models. N=4113

Method	Number of AK's	Unadjusted OR (95% CI)	Adjusted* OR (95% CI)	Fully adjusted** OR(95% CI)
Binary logistic regression	AK ≥1	0.60 (0.42-0.83)	0.74 (0.51-1.05)	0.69 (0.47-1.07)
Multinomial logistic regression* **	AK 1-3	0.75 (0.51-1.12)	0.89 (0.60-1.33)	1.12 (0.64-1.96)
	AK 4-9	0.56 (0.28-1.11)	0.75 (0.37-1.50)	0.75 (0.27-2.08)
	AK ≥10	0.20 (0.07-0.64)	0.28 (0.09-0.90)	0.19 (0.03-1.43)

*adjusted for gender and age

**adjusted for gender, age, pigmentation status, history of living in a sunny country, sun protective behaviour, male baldness, number of sunburns, smoking and use of immunosuppressive medication

*** reference category 0 AK, p<0.001 for trend in decreasing ORs across AK categories

In order to determine which of the AD criteria were driving the association (barrier defect or chronic inflammation), we analysed the criteria separately. From the total group of 260 persons diagnosed with AD, 92 had active dermatitis, in 209 there was flexural involvement, 169 had a history of atopy and 119 were diagnosed with eczema before the age of 12. The (un)adjusted odds ratios for these criteria are shown in table 3. Two minor criteria, active dermatitis and atopy in history had a significant effect on the AD-AK association adjusted OR 0.57; 95% CI, 0.42-0.77 and 0.75; 95%CI, 0.60-0.92 respectively. Exclusion of hand dermatitis from the analysis did not alter the OR essentially, however the CI became insignificant due to loss of power OR 0.62; 95% CI 0.25-1.55 and aOR 0.70; 95%CI (0.27-1.84).

Table 3. Binary logistic regression models (unadjusted/adjusted) for the association between AK and separate AD-criteria

Variable	Cases (N)	Controls (N)	Unadjusted OR (95% CI)	Adjusted* OR (95% CI)
AD criteria (all)	42	218	**0.60 (0.42-0.83)**	0.74 (0.51-1.05)
Hand dermatitis excluded	34	170	0.62 (0.25-1.55)	0.70 (0.27-1.84)
Active dermatitis	60	292	**0.63 (0.47-0.84)**	**0.57 (0.42-0.77)**
Atopy in history	142	608	**0.62 (0.51-0.76)**	**0.75 (0.60-0.92)**
Eczema diagnosed <12 years	49	221	**0.73 (0.53-1.00)**	0.91 (0.65-1.27)
Flexural itch in history	101	407	0.84 (0.66-1.06)	0.98 (0.76-1.26)

* adjusted for gender and age

AD and keratinocyte cancers

Of the 4.375 people in the RS, 350 (8%) had one or more BCCs of whom 16 persons had AD and 336 did not have AD (p=0.100). In binary logistic regression having AD did not increase the likelihood of having a BCC with and without adjustment for age and gender (OR 0.64; 95% CI, 0.37-1.11 and adjusted OR 0.71; 95% CI, 0.41-1.24). Seventy-eight persons had one or more SCCs. Six of them had AD and 72 did not meet the criteria for AD (p=0.500). AD did not change the likelihood of having a SCC with an unadjusted odds ratio 1.33 (95% CI, 0.57-3.08) and adjusted OR1.54 (95% CI, 0.66-3.62).

DISCUSSION

Our findings suggest that within a population based sample, AD patients do not have more AKs than the rest of the population. Moreover, this study indicates that in individuals with AD the odds of having more than10AKs are significantly lower. A gradient of decreasing ORs with the increasing number of AKs was discernible suggesting a biological effect. We did not find significant associations between KCs (both BCC and SCC) and atopic dermatitis. However, this study was not powered for the subanalyses and for the association between AD and BCC and SCC. Nonetheless, the results of our study suggest that the average, community-dwelling AD-patients are not at an increased risk of AK and keratinocyte cancers.

In the past, two hypotheses explained the associations between AD and KC. One dictates that the overactive, atopic immune system in AD brings about increased immunosurveillance of neoplastic cells while the other implies that the impaired epidermal barrier of AD actually predisposes to more UV-induced damage and subsequent KC.[3]

When analysing the AD criteria separately it appears that the minor criterion 'active dermatitis' is the strongest driver of the negative association suggesting that patients with long-standing and/or active AD are more protected . Actinic keratoses are usually accompanied by erythema and inflammation which can be visualised by histopathology. This inflammatory response can lead to spontaneous remission of immunogenic AKs. Eczematous skin may mount this inflammatory response more easily.[7] An alternative explanation for the protective effect we have found may be the locally increased turn-over of the keratinocytes or altered signalling between keratinocytes and immune cells.[6,30] A cohort-study of 57815 patients who had been tested for total serum IgE and Phadiatop revealed no effect of serological atopy on the incidence of various cancers including KCs. This suggests no general protective effect of systemic atopy.[31] It is also possible that a difference in behaviour between AD patients and control could explain the difference. For example, AD-patients may be more aware of their skin and sun damage and may be more adherent to sun protective behaviours. There is a possibility of detection bias if patients with AD present to physicians more often and have their AKs treated. Furthermore, AD patients may be more skilled in the correct and frequent application of sunscreens or may avoid sun in general because of embarrassment with skin lesions.

This is the first study to examine the association between AD and AK. The strengths of this study are the large number of patients, the fact that the presence of AK was diagnosed by trained physicians and that the number of AKs was established in detail. Furthermore we took into account several confounders, which enhances the validity of the study. There are some limitations to this study. This was a cross-sectional study. The outcome (AK) has been determined by a single measurement without longitudinal data. AK and AD have very differing ages of peak prevalence. In fact, the development of AD may be preceded by the development of AK by decades which makes a longitudinal study starting at age of onset of AD during up to the age of 40 when first AKs appear impossible at this moment. This necessitates retrospective ascertainment of exposure / AD to some degree. There are no validated AD diagnostic criteria for adults. The use of modified UK working party´s criteria may have led to the inclusion of other forms of eczema. This would lead to a dilution of effect. The group which met the AD criteria comprised 6.9% of the total study population. This is comparable to the prevalence of AD in children from the 40's and 50's (childhood years in our study population), as described in literature.[32] Moreover, the exclusion of hand dermatitis in our sensitivity analyses did not lead to different results. Unfortunately we could not assess the overlap in skin areas for eczematous skin and actinic keratoses. In this study we did not know which patients received treatment for severe AD like immunosuppressants and UVB therapy. These treatments contribute to the risk of cutaneous (pre)malignancies and may have led to uncontrolled confounding of AD on AK and KC risk and could explain why some

of the studies in AD-cohorts have found an increased risk. However, our AD-patients are community dwelling and the percentage of severe AD treated by these modalities is expected to be low. Several previous studies have also found an inverse relationship between atopic dermatitis and KC. One case-control study showed an adjusted OR 0.78; 95% CI, 0.61-0.98, but the authors used subjects with other cutaneous diseases as the control group, which might have influenced their results.[23] Margolis et al. found similar results, but selected their controls out of a 'dermatitis' group. Kaae et al. examined the filaggrin gene mutation status and found non-significant odds ratios below 1.00. They concluded that filaggrin mutation carrier-status is not important for the association between AD and BCC.[33]

However the pathomechanism remains unclear. Future studies should focus on physician-made diagnosis of AD as well as on the filaggrin gene status as exposures. It is also necessary to examine if the sites of KCs and AKs overlap with the skin areas affected by atopic dermatitis.

CONCLUSION

Patients with AD were not found to have more of AKs or KCs. Moreover, individuals with atopic dermatitis seem to be less likely to develop multiple AKs.

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REFERENCES

1. Williams H, Flohr C. How epidemiology has challenged 3 prevailing concepts about atopic dermatitis. *J Allergy Clin Immunol* 2006; **118**: 209-13.
2. Bieber T. Atopic dermatitis. *N Engl J Med* 2008; **358**: 1483-94.
3. Hajdarbegovic E, Verkouteren J, Balak D. Non-melanoma skin cancer: the hygiene hypothesis. *Med Hypotheses* 2012; **79**: 872-4.
4. Mildner M, Jin J, Eckhart L *et al.* Knockdown of filaggrin impairs diffusion barrier function and increases UV sensitivity in a human skin model. *J Invest Dermatol* 2010; **130**: 2286-94.

5. Denecker G, Hoste E, Gilbert B *et al.* Caspase-14 protects against epidermal UVB photodamage and water loss. *Nat Cell Biol* 2007; **9**: 666-74.
6. Guinea-Viniegra J, Zenz R, Scheuch H *et al.* Differentiation-induced skin cancer suppression by FOS, p53, and TACE/ADAM17. *J Clin Invest* 2012; **122**: 2898-910.
7. Strid J, Sobolev O, Zafirova B *et al.* The intraepithelial T cell response to NKG2D-ligands links lymphoid stress surveillance to atopy. *Science* 2011; **334**: 1293-7.
8. Dupuy A, Dehen L, Bourrat E *et al.* Accuracy of standard dermoscopy for diagnosing scabies. *J Am Acad Dermatol* 2007; **56**: 53-62.
9. Madan V, Lear JT, Szeimies RM. Non-melanoma skin cancer. *Lancet* 2010; **375**: 673-85.
10. He W, Zhu F, Ma X *et al.* Actinic skin damage and mortality--the First National Health and Nutrition Examination Survey Epidemiologic Follow-up Study. *PLoS One* 2011; **6**: e19907.
11. Flohil SC, van der Leest RJ, Dowlatshahi EA *et al.* Prevalence of actinic keratosis and its risk factors in the general population: the Rotterdam Study. *J Invest Dermatol* 2013; **133**: 1971-8.
12. El-Khateeb EA, Ragab NF, Mohamed SA. Epidermal photoprotection: comparative study of narrowband ultraviolet B minimal erythema doses with and without stratum corneum stripping in normal and vitiligo skin. *Clin Exp Dermatol* 2011; **36**: 393-8.
13. Hong SP, Kim MJ, Jung MY *et al.* Biopositive effects of low-dose UVB on epidermis: coordinate upregulation of antimicrobial peptides and permeability barrier reinforcement. *J Invest Dermatol* 2008; **128**: 2880-7.
14. Barresi C, Stremnitzer C, Mlitz V *et al.* Increased sensitivity of histidinemic mice to UVB radiation suggests a crucial role of endogenous urocanic acid in photoprotection. *J Invest Dermatol* 2011; **131**: 188-94.
15. Skaaby T, Husemoen LL, Thyssen JP *et al.* Filaggrin loss-of-function mutations and incident cancer: a population-based study. *Br J Dermatol* 2014; **171**: 1407-14.
16. Dyer RK, Weinstock MA, Cohen TS *et al.* Predictors of basal cell carcinoma in high-risk patients in the VATTC (VA Topical Tretinoin Chemoprevention) trial. *J Invest Dermatol* 2012; **132**: 2544-51.
17. Arana A, Wentworth CE, Fernandez-Vidaurre C *et al.* Incidence of cancer in the general population and in patients with or without atopic dermatitis in the U.K. *Br J Dermatol* 2010; **163**: 1036-43.
18. Jensen AO, Svaerke C, Kormendine Farkas D *et al.* Atopic dermatitis and risk of skin cancer: a Danish nationwide cohort study (1977-2006). *Am J Clin Dermatol* 2012; **13**: 29-36.
19. Hagstromer L, Ye W, Nyren O *et al.* Incidence of cancer among patients with atopic dermatitis. *Arch Dermatol* 2005; **141**: 1123-7.
20. Hwang CY, Chen YJ, Lin MW *et al.* Cancer risk in patients with allergic rhinitis, asthma and atopic dermatitis: a nationwide cohort study in Taiwan. *Int J Cancer* 2012; **130**: 1160-7.
21. Milan T, Verkasalo PK, Kaprio J *et al.* Lifestyle differences in twin pairs discordant for basal cell carcinoma of the skin. *Br J Dermatol* 2003; **149**: 115-23.
22. Cheng J, Zens MS, Duell EJ *et al.* History of Allergy and Atopic Dermatitis in Relation to Squamous Cell and Basal Cell Carcinoma of the Skin. *Cancer Epidemiol Biomarkers Prev* 2015.
23. Ming ME, Levy R, Hoffstad O *et al.* The lack of a relationship between atopic dermatitis and non-melanoma skin cancers. *J Am Acad Dermatol* 2004; **50**: 357-62.
24. Hofman A, Darwish Murad S, van Duijn CM *et al.* The Rotterdam Study: 2014 objectives and design update. *Eur J Epidemiol* 2013; **28**: 889-926.
25. Bolognia J, Jorizzo J, Schaffer J. In: *Dermatology: 2-Volume Set, 3rd Edition*. 2012; 203-16.
26. Paul C, Maumus-Robert S, Mazereeuw-Hautier J *et al.* Prevalence and risk factors for xerosis in the elderly: a cross-sectional epidemiological study in primary care. *Dermatology* 2011; **223**: 260-5.
27. Jacquet A. Innate immune responses in house dust mite allergy. *ISRN Allergy* 2013; **2013**: 735031.

28. Zeppa L, Bellini V, Lisi P. Atopic dermatitis in adults. *Dermatitis* 2011; **22**: 40-6.
29. Wakkee M, Hollestein LM, Nijsten T. Multivariable analysis. *J Invest Dermatol* 2014; **134**: e20; quiz e.
30. Cipolat S, Hoste E, Natsuga K *et al.* Epidermal barrier defects link atopic dermatitis with altered skin cancer susceptibility. *Elife* 2014; **3**: e01888.
31. Lindelof B, Granath F, Tengvall-Linder M *et al.* Allergy and cancer. *Allergy* 2005; **60**: 1116-20.
32. Taylor B, Wadsworth J, Wadsworth M *et al.* Changes in the reported prevalence of childhood eczema since the 1939-45 war. *Lancet* 1984; **2**: 1255-7.
33. Kaae J, Thyssen JP, Johansen JD *et al.* Filaggrin Gene Mutations and Risk of Basal Cell Carcinoma. *Br J Dermatol* 2013.

Chapter 3.3

Atopic dermatitis is not a protective factor for melanoma but asthma may be.

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ABSTRACT

Background: There is evidence from cohort studies for an inverse association between atopic dermatitis and asthma and cutaneous melanoma. However, these studies have been too heterogeneous and did not show statistically significant results. Also, this association has not been compared to traditional melanoma risk factors.

Objectives: To test for associations between history of atopic disorders and melanoma life time prevalence. To test for associations between atopic disorders and melanoma prognosis.

Methods: Validated questionnaires from the European Community Respiratory Health Survey and International Study of Asthma and Allergies in Children protocol on life time prevalence of atopic disorders were sent to 280 patients with histopathologically confirmed melanoma. The control group consisted of their spouses. Also the skin phototype was assessed using a validated questionnaire.

Results: One hundred and eighty four melanoma patients and 169 controls responded to the questionnaire. The life time prevalence of atopic dermatitis and hayfever was not different in melanoma patients(8.7% vs. 8.2 p=0.890 and 15.2% vs. 18.3% p=0.432 respectively). Asthma was non-significantly lower in melanoma patients (3.8% vs. 8.2% p=0.075). Atopic melanoma patients did not differ from non-atopic patients in terms of Breslow thickness, metastases and second melanomas.

Conclusion: Atopic dermatitis is not a protective factor in cutaneous melanoma but a history of asthma may be.

BACKGROUND

Cutaneous melanoma is one of the most aggressive malignant tumours and is virtually incurable after metastasis. It is known for its rapid growth and the ability to metastasize early via the lymphatics and the blood vessels. The incidence of melanoma in US is 3.0 and in Europe it is 2.2-19.2 in 100.000 and it has been rising in the past 30 years. [1,2] The conventional risk factors for development of melanoma are fair skin, history of melanoma in the index patient or their family members, having clinically atypical moles, sunburns and exposition to UV-radiation.[3, 4] Because the knowledge of the traditional risk factors has not lead to effective prevention programs it is worthwhile looking into alternative risk factors.[5] Atopic disorders; atopic dermatitis, hayfever and atopic asthma have been of interest in epidemiological studies as a risk and a protective factor for cancer. These studies have shown lower prevalences of different types of cancer in atopic individuals.[6, 7] In two cohort studies with more than 31000 patients with atopic eczema a decreased prevalence of MM, albeit borderline significant, has been found with a total of 6 and 12 melanoma patients respectively.[8,9] In three prospective studies children with atopic dermatitis had significantly fewer nevi which was implied to reduce the risk of melanoma.[10-12] On the other hand a health survey in 3.308 Australians revealed an increased prevalence of melanoma (27 cases with hazard ratio 2.39, 95% CI [1.00-5.70]) in men with hayfever.[13] In contrast, another German case-control study from 1988 with 331 melanoma patients demonstrated that melanoma patients were less likely to report atopic symptoms (4.2% vs. 10%). [14] The effect of asthma has also been assessed in a cohort of hospitalized asthmatics. The standardized incidence of melanoma was found to be decreased in a ten-year period in 1970s in Sweden, but only 72 melanoma cases occurred.[15] This was also found in another large cohort study of same methodology with 64 346 asthma patients.[16] However this finding was not consistent for all decades and was not found in patients with multiple admissions for asthma. These previous studies have been heterogeneous, registry-based and focussing on a single atopic disorder while assessing the incidences of multiple cancers simultaneously. From their data we can not conclude weather atopic disorders are a risk factor for development or progression of cutaneous melanoma.

PATIENTS AND METHODS

Study population

The melanoma patients were recruited from the department of Dermatology and Venereology of the Erasmus Medical Centre in Rotterdam. They comprised 280,Dutch, Caucasian patients diagnosed with a cutaneous melanoma between 2000 and 2010

extracted from our administrative registry. The diagnosis was histologically confirmed in all patients by certified pathologists. The control group consisted of melanoma patient´s spouses. All subjects were sent identical questionnaires concerning atopic manifestations and their treatment. Melanoma patients and their spouses were also sent a questionnaire assessing their skin phototype. The choice for this control group was made because of availability and because of the match in ethnic, socio-economic and external factors without genetic bias. Because atopic disorders are largely genetically determined and are usually seen in childhood it is not to be expected that this exposure is of any influence on partner choice later in life. The medical ethical committee at Erasmus Medical Center provides a waiver for single-subject questionnaire based studies. This study has been conducted in accordance with the Declaration of Helsinki.

Patient reported atopic disease and skin type

The subjects were approached through a postal survey. Non-responders were contacted a second time. They were sent a questionnaire comprising questions from the European Community Respiratory Health Survey and International Study of Asthma and Allergies in Children protocol.[17, 18] Subjects were considered to have asthma if they positively answered the question "Have you ever been diagnosed with asthma by a doctor?" and if the answer to question "How old were you when you were diagnosed with asthma?" was less than 25 years in order to prevent misclassification of subjects with chronic obstructive pulmonary disease. Diagnosis of atopic dermatitis was made in patients with eczema diagnosed by a physician, with an onset before age 25 and duration of more than 1 year. For diagnosis of hayfever patients had to answer next question affirmatively; "Have you ever had hayfever?" and they would have to have it before the age of 25. Because atopic disorders wane after childhood we have deemed patients diagnosed before the age of 25 to be atopic. This also prevents inclusion of chronic pulmonary obstructive disease patients as asthmatics. Also other forms of dermatitis are more likely after the age of 25. To assess the Fitzpatrick skin type the patients filled in the `Fitzpatrick skin type questionnaire.[19] This allows the comparison of the effect of atopy and a conventional risk factor which is a skin phototype lower than 3.

Statistics

For sample size calculations, we assumed a prevalence of 20% of atopic disorders in our control group, as we have found previously[20] and expected half of this proportion in melanoma patients.[14] For a power of 80% and an alpha level of 0.05, a sample size of 135 cases and 135 controls was required. The Chi-squared test was used to test for statistical differences in proportions. The logistic multiple regression analysis was performed in standard fashion where all variables with $p<0.20$ from the univariate analysis were included. In order to be able to assess and compare the effect size of individual

determinants, the variables have analyses by enter method. All data were analysed using IBM SPSS version 19.0 (Chicago, Illinois 2011)

RESULTS

Demographic characteristics

A total of 184(61%) Dutch melanoma patients and 169(56%) of their spouses responded to the questionnaire. (Table 1) The control subjects were younger (53 vs. 57 years p=0.001) and more frequently male (53% vs. 41% p=0.001). The family history of atopy did not differ across the groups. Melanoma patients had a skin phototype lower than 3 more frequently than controls (60% vs. 36% p<0.001). The median Breslow-thickness of our patients was 1.1 mm [IQR 0.66-2.00]. Twenty-three of them had metastases and 29 had a second melanoma. The two control group components did not differ with regard to demographic characteristics or atopic features.

Table 1. Subject characterisitcs

	Melanoma patients	Controls	p-value
Number	184	169	
Mean age (SD)	57 (14)	56 (14)	0.274
Male sex	75 (41%)	86 (51%)	0.056
Family history of atopy	95 (52%)	79 (47%)	0.359
Skin phototype < 3	108 (60%)	60 (36%)	<0.001
Breslow thickness median (mm)	1,1 IQR[0.66-2.00]	-	-
Metastases	23 (13%)	-	-
Second melanoma	29 (16%)	-	-

Abbreviations: SD, standard deviation; IQR, interquartile range
Definitions: Metastases; lymphatic or distant site metastases, Second melanoma; new primary cutaneous melanoma

Atopic features

There were no statistically significant differences in atopic eczema, hayfever and the use of disease specific medication across the groups.(Table 2). Melanoma patients reported less frequently to have asthma but this was not statistically significant (3.8% vs. 8.2% p=0.075). In the univariate regression analysis the strongest association was found between melanoma and high risk skin phototype i.e. skin phototypes 1 and 2 (OR 2.69, 95% CI[1.74-4.14]) (Table 3). Nevertheless, history of asthma seemed to be inversely associated with melanoma OR 0.44 95% CI (0.17-1.11) but this was not statisti-

cally significant. High risk skin phototype remained the most important risk factor in the multiple regression analysis adjusted OR 2.60. 95% CI [1.66-4.05].

Table 2. Life time prevalence of atopic features in 184 melanoma patients versus 169 controls

	Melanoma patients	Controls	p-value
Number	184	169	
Asthma	7 (3.8%)	14 (8.2%)	0.075
Current inhalator use	7 (3.8%)	11 (6.5%)	0.249
Atopic dermatitis	16 (8.7%)	14 (8.2%)	0.890
Current topical corticosteroid use	26 (14.1%)	19 (11.2%)	0.416
Hayfever	28 (15.2%)	31 (18.3%)	0.432
Current use of antihistaminics	22 (12.0%)	23 (13.6%)	0.642
Any atopic disorder	44 (24.0%)	45 (26.6%)	0.557

Table 3. Standard uni- and multivariate regression analysis for risk of melanoma

	Unadjusted OR for melanoma with 95% CI	Adjusted OR for melanoma with 95% CI
Age	**1.01 (1.00-1.03)**	**1.01 (1.0-1.03)**
Male sex	**0.66 (0.44-1.01)**	0.72 (0.46-1.12)
Skin phototype < 3	**2.69 (1.74-4.14)**	**2.61 (1.67-4.07)**
Asthma	**0.44 (0.17-1.11)**	0.53 (0.20-1.41)
Atopic dermatitis	1.05 (0.50-2.23)	**n/a**
Hayfever	0.80 (0.46-1.40)	**n/a**
Any atopic disorder	0.87 (0.54-1.40)	**n/a**

Variables with p > 0.20 were left out from multivariate analysis. Variables with p<0.20 are printed in bold.
Abbreviations: OR; odds ratio, 95% CI; 95% confidence intervals

Atopy in melanoma patients

For the analysis of the effect of atopy on prognostic parameters of melanoma we compared melanoma patients with, eczema, hayfever or asthma to patients who reported none of these atopic disorders. Atopic melanoma patients were younger on the average than the non-atopic patients (54 vs. 59 years, p=0.035) (Table 4). Nevertheless, atopic patients did not differ from non-atopic patients in terms of Breslow-thickness, metastases and second melanomas. However, high risk skin types were more prevalent in atopic patients (70% vs. 57% p=0.085). The seven asthmatic melanoma patients did not differ from the non-asthmatic in terms of Breslow thickness 0.9 IQR[0.4-2.4] vs. 1.1 IQR[0.7-2.0]. Grouping the patients according to atopic dermatitis and hayfever status yielded essentially same results.

Table 4. Atopic versus non-atopic melanoma patients

	Atopic melanoma patients	Non-atopic melanoma patients	p-value
Number of patients	44	140	
Age, mean (SD)	54(14)	59(14)	P=0.035
Age at diagnosis, mean (SD)	48(15)	52/(15)	P=0.309
Male sex	16/44	59/140	P=0.309
Skin phototype < 3	30/43	78/138	P=0.085
Breslow thickness, median [IQR]	1.00 (0.51-2.02)	1.10 [0.70-2.00]	P=0.722
Metastases	5/44	18/140	P=0.514
Second melanoma	8/44	21/140	P=0.384

Abbreviations: SD, standard deviation; IQR, interquartile range
Definitions: Metastases; lymphatic or distant site metastases, Second melanoma; new primary cutaneous melanoma

DISCUSSION

Our findings indicate that atopic dermatitis and hayfever are not a protective or a risk factor for melanoma. Asthma however may be associated with a lower risk of melanoma as found in previous studies.[15, 16] Although they were more prevalent, hayfever and atopic eczema did not show any association with melanoma. In comparison, having a high risk skin phototype was found to be significantly associated with melanoma and its effect on the odds ratio was much greater than the effect of having an atopic disorder. Atopic disorders were also not associated with prognostic factors for melanoma like Breslow thickness and metastasis. Greatest limitation of our study is the low number of cases although our study sufficiently excludes a large effect from atopic dermatitis and hayfever. A bigger study is necessary to confirm the findings in patients with asthma. Also the retrospective setup may introduce selection bias because of the melanoma lethality. However, a case control methodology is preferred because of the high prevalence of atopic disorders i.e. exposure and relatively low prevalence of melanoma. Also, there is not a more practical way to ascertain atopy as it is mostly found during childhood. It is noteworthy that atopic melanoma patients had lower skin phototypes than non-atopic patients which is actually expected to increases their chance of having a melanoma. It also means that adjustment for skin phototype is warranted in future studies. Because of the different skin phototype in controls there is a possibility of different ethnicities which may also influence the prevalence of atopy. There is no obvious mechanism to explain how asthma may lower the risk of melanoma. We suggest less exposure to sun because of less outdoor activities or an altered immune system be looked into in future studies. Traditional risk factors for melanoma such as the skin phototype and number of nevi should be also taken into account

when assessing alternative risk factors. We believe that our study is of value for future investigations, suggesting a methodological approach. For now, our data to suggest that asthma may protect from cutaneous melanoma.

REFERENCES

1. Forsea, A.M., et al., *Melanoma incidence and mortality in Europe: new estimates, persistent disparities.* Br J Dermatol.
2. Ward, E.M., et al., *Interpreting cancer trends.* Ann N Y Acad Sci, 2006. **1076**: p. 29-53.
3. Moan, J., A.C. Porojnicu, and A. Dahlback, *Ultraviolet radiation and malignant melanoma.* Adv Exp Med Biol, 2008. **624**: p. 104-16.
4. Dennis, L.K., et al., *Sunburns and risk of cutaneous melanoma: does age matter? A comprehensive meta-analysis.* Ann Epidemiol, 2008. **18**(8): p. 614-27.
5. Diepgen, T.L. and V. Mahler, *The epidemiology of skin cancer.* Br J Dermatol, 2002. **146 Suppl 61**: p. 1-6.
6. El-Zein, M., et al., *History of asthma or eczema and cancer risk among men: a population-based case-control study in Montreal, Quebec, Canada.* Ann Allergy Asthma Immunol, 2010. **104**(5): p. 378-84.
7. Merrill, R.M., R.T. Isakson, and R.E. Beck, *The association between allergies and cancer: what is currently known?* Ann Allergy Asthma Immunol, 2007. **99**(2): p. 102-16; quiz 117-9, 150.
8. Synnerstad, I., et al., *Low risk of melanoma in patients with atopic dermatitis.* J Eur Acad Dermatol Venereol, 2008. **22**(12): p. 1423-8.
9. Jensen, A.O., et al., *Atopic dermatitis and risk of skin cancer: a Danish nationwide cohort study (1977-2006).* Am J Clin Dermatol, 2012. **13**(1): p. 29-36.
10. Kallas, M., et al., *Frequency and distribution pattern of melanocytic naevi in Estonian children and the influence of atopic dermatitis.* J Eur Acad Dermatol Venereol, 2006. **20**(2): p. 143-8.
11. Broberg, A. and A. Augustsson, *Atopic dermatitis and melanocytic naevi.* Br J Dermatol, 2000. **142**(2): p. 306-9.
12. Synnerstad, I., et al., *Fewer melanocytic nevi found in children with active atopic dermatitis than in children without dermatitis.* Arch Dermatol, 2004. **140**(12): p. 1471-5.
13. Talbot-Smith, A., et al., *Allergy, atopy, and cancer: a prospective study of the 1981 Busselton cohort.* Am J Epidemiol, 2003. **157**(7): p. 606-12.
14. Kolmel, K.F. and D. Compagnone, *[Melanoma and atopy].* Dtsch Med Wochenschr, 1988. **113**(5): p. 169-71.
15. Ji, J., et al., *Cancer risk in hospitalised asthma patients.* Br J Cancer, 2009. **100**(5): p. 829-33.
16. Kallen, B., J. Gunnarskog, and T.B. Conradson, *Cancer risk in asthmatic subjects selected from hospital discharge registry.* Eur Respir J, 1993. **6**(5): p. 694-7.
17. Asher, M.I., et al., *International Study of Asthma and Allergies in Childhood (ISAAC): rationale and methods.* Eur Respir J, 1995. **8**(3): p. 483-91.
18. Burney, P.G., et al., *The European Community Respiratory Health Survey.* Eur Respir J, 1994. **7**(5): p. 954-60.
19. Fitzpatrick, T.B., *The validity and practicality of sun-reactive skin types I through VI.* Arch Dermatol, 1988. **124**(6): p. 869-71.
20. Hajdarbegovic, E., et al., *Decreased prevalence of atopic features in patients with psoriatic arthritis, but not in psoriasis vulgaris.* J Am Acad Dermatol, 2012.

Chapter 4

Atopy and cardiovascular disease

Chapter 4.1

Increased prevalence of coronary heart disease in older atopic dermatitis patients

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Submitted

ABSTRACT

Importance: Previous research has suggested an association between atopic dermatitis and coronary heart disease. These studies did not employ validated measures of cardiovascular disease, its risk factors and validated diagnostic criteria for atopic dermatitis and/or included mostly hospitalized AD patients.

Objective: The objective of this study was to assess the associations of coronary heart disease, its risk factors and atopic dermatitis with validated disease definitions and prospectively gathered, predefined data from a population based cohort.

Design: We performed cross sectional analyses of the data from the prospective cohort, the Rotterdam Study. Multivariable models were constructed taking into account all traditional cardiovascular risk factors.

Setting: Population based.

Participants: Inhabitants of the Ommoord district in Rotterdam.

Exposures: Atopic dermatitis was defined according to the modified UK working party's criteria.

Main Outcome Measures: The coronary heart disease status was obtained through prospective checks of all patients' records, regular medical checks, laboratory and radiological investigation, electrocardiograms, home interviews and municipality records.

Results: We screened 4386 subjects of whom 723 were found to have AD. There were 63 cases of coronary heart disease in the atopic dermatitis group and 276 in the control group (8.7% vs 7.6%, p=0.358)
After adjusting for sex and age an OR 1.42 95%CI [1.05-1.89] was found. In the fully adjusted model this was OR 1.54 95%CI [1.09-2.15]. Atopic dermatitis did not associate positively with any of the traditional cardiovascular risk factors.

Conclusion: Atopic dermatitis in community dwelling patients is associated with coronary heart disease but it is this association is independent of the traditional cardiovascular risk factors.

KEY POINTS

Question: Is atopic dermatitis associated with coronary heart disease in community dwelling patients?

Findings: Elderly AD patients are more likely to have had coronary heart disease than controls after adjusting for traditional cardiovascular risk factors.

Meaning: The association between atopic dermatitis and coronary heart disease is independent of the traditional risk factors in community dwelling patients.

ABBREVIATIONS

AD; Atopic dermatitis
CHD; coronary heart disease
CVD; cardiovascular disease
OR; Odds ratio
RS; Rotterdam study
95% CI; 95% confidence interval

INTRODUCTION

Atopic dermatitis (AD) is one of the most prevalent inflammatory skin conditions affecting up to 30% of infants and about 10% of adults and its incidence has tripled in the past three decades. [1]Atopic dermatitis is the result of an impaired epidermal barrier, partly due to mutations in the filaggrin gene, combined with an overactive, Th2-polarized immune system. [2]

Traditionally, associated disorders i.e. comorbidities are atopic asthma, food allergies and allergic rhino-conjunctivitis. However in dermatology journals increasing attention is being given to systemic effects and cardiovascular comorbidities in patients with AD in parallel with extensively studied psoriasis vulgaris, another prevalent inflammatory dermatosis.[3] From the cardiologist point of view there is a need for non-laboratory, clinically identifiable indicators of risk of cardiovascular disease (CVD) such as skin signs.[4] Previous studies on this matter either used a self-reported definition of AD and cardiovascular risk factors, focused on risk factors rather than cardiovascular events or were based on administrative health insurance data and did not fully control for known CVD risk factors and/or included exclusively hospitalized AD patients.

In a recent U.S. population-based study by Silverberg et al, self-reported AD patients were found to be more likely to be exposed to cardiovascular risk factors such as smoking (aOR, 1.28; 95% CI, 1.12-1.45), drinking of alcoholic beverages (aOR, 1.16; 95% CI, 1.03-1.31), having higher body mass index (BMI) (adjusted β, 0.86; 95% CI, 0.37-1.36) and hypertension (aOR, 1.56; 95% CI 1.22-1.99).[5] Another population-based study by the same team found increased prevalences of self-reported acute myocardial infarction (MI), angina and peripheral vascular disease (PVD) in the U.S. [6] The Nurses' Health Study, also in U.S., with 78,702 participants found no association between self-reported AD and non-fatal MI.[7] They did not report specifically on the associations between AD and CVD risk factors. A Taiwanese population-based study of health care administrative data of 20,323 AD patients showed and increased incidence rate (IR, 1.33; 95%CI, 1.12-1.59) of ischemic stroke compared with co-morbidity matched controls in Thailand.[8] This effect was stronger in patients with severe atopic dermatitis. In contrast, in the studies of European populations no association between AD and CVD risk factors was detected according to a review from 2015.[9] However, a recent study of an administrative cohort of 1.2 million subjects in Germany revealed an adjusted relative risk of (aRR 1.32; 95% CI 1.26-1.38) of angina pectoris (AP) in AD patients.[10] In addition to the aforementioned variations in outcomes, a Danish study of administrative health care data revealed a protective effect of mild AD, defined as AD without systemic treatment, on coronary heart disease (IRR 0.73; 95% CI 0.59-0.91).[11] In this study severe AD was not significantly

associated with coronary heart disease in age and sex and fully adjusted analyses (IRR 1.39; 95%CI 0.95-2.03).

The objective of our study was to investigate whether AD, diagnosed with modified U.K. Working Party's diagnostic criteria associates with coronary heart disease (CHD) and CVD risk factors within a population-based Dutch cohort (i.e. Rotterdam Study) with detailed and validated information available on CHD, CVD risk factors and AD. [12]

METHODS

Study design and study population

The Rotterdam study (RS) is an ongoing prospective population-based cohort study that follows inhabitants of the Ommoord district of Rotterdam, the Netherlands since 1990.[12] It is designed to make possible high standard research of CVD as its primary objective. In January 1990, the first cohort (RS-I) of 7,938 participants (78% of invitees) aged 55 years or older was established. In 2000, a second cohort (RS-II) was added to the RS, including 3,011 participants (67% of invitees) who had turned 55 years of age or had moved into the study district. The third cohort (RS-III) was established in 2006, in which 3,932 participants (65% of invitees) aged 45-54 years were added to the cohort. The RS reflects the population of the Netherlands in terms of co-morbidities and health consumption. Participants of the present study were all above 50 years of age and >95% are of Caucasian descent. Dermatologic screening was introduced in August 2010. Since then, full body surface examinations (FBSE) (with the exception of the skin covered by socks and underwear) are being conducted by four dermatologists trained physicians focusing on the most common skin diseases such as skin (pre-)malignancies, atopic dermatitis, hand eczema, psoriasis, and varicose veins. All participants of the RS are dermatologically screened. The current study uses data from the extended cohorts comprising subjects who have been dermatologically screened. It is a cross-sectional analysis of the currently available RS data. The RS is approved by the Medical Ethics of the Erasmus MC, University Medical Center and The Netherlands Ministry of Health and is conducted in concordance with the Helsinki Guidelines. All participants in the study gave written informed consent.

Exposure: Atopic dermatitis

The participants were interviewed at home and full body surface examination (FBSE) was performed by dermatologist trained physicians in a medical research facility in the centre of the RS district between 2010 and 2015. The FBSE was structured by a predefined set of common dermatologic findings and their localizations such as actinic

keratosis, basal cell carcinoma, eczema, psoriasis etc. The research physicians as well as the participants were unaware of the current hypothesis. The diagnosis of AD was made according to the diagnostic guidelines for atopic dermatitis from the U.K. working party which is the most validated diagnostic tool in the clinical and investigational settings.[13] These criteria comprise one major criterion plus three or more minor criteria. 'An itchy skin condition in past 12 months' is the must have criterion and the participant needs to fulfil at least three minor criteria which are: 'history of involvement of the skin creases', 'a personal history of asthma or hay fever', 'a history of generally dry skin in the last year', 'visible flexural eczema' and 'onset under 2 years of age'. However, no diagnostic criteria of AD have been validated for adult populations such as the RS. We therefore made adjustments to the U.K. working party's criteria prior to the analyses based on our previous publication.[14] To include patients with current remission another criterion was added "diagnosis of eczema by a physician ever". The personal history of other atopic diseases was extended with 'house dust mite allergy', because it is strongly related to atopic diseases in adults.[15] Furthermore sites of visible eczema were adapted. In adults, atopic dermatitis also occurs on the extremities, hands and trunk.[16] Finally the onset age was prolonged to ever instead of 2 years of age, to include patients with later onsets.[16] Thus, patients who reported having an itchy condition and who fulfilled at least 3 of the other 5 criteria were considered to have AD in this study. Patients who did not fulfil these criteria were considered unexposed.

Outcomes: CHD and CVD risk factors

The outcomes in this study is prevalent coronary heart disease and has been described in detail previously.[17] In short, coronary heart disease (CHD) was defined as fatal or non-fatal myocardial infarction, surgical or percutaneous coronary revascularisation procedure (as a proxy for unstable or incapacitating angina), or death from coronary heart disease. RS has been specifically designed with the objective of studying CHD comprising MI, myocardial revascularization, mortality through CHD, and heart failure. This is established through regular checks of medical records at the family doctor's office, continuous linkage with electronic patient's records, laboratory and radiologic investigation, electrocardiograms home interviews, hospital discharge letters, pharmacy prescription records, and municipality records.

Covariates and cardiovascular risk factors

We considered all traditional risk factors of CHD as covariates in our fully adjusted multivariable models. These are: age, sex, smoking status as assessed by multiple home interviews, body mass index (BMI), total serum cholesterol, serum high-density lipoprotein, systolic and diastolic blood pressures, medication for hyperlipidemia, diabetes mellitus defined as use of oral glucose lowering drugs or insulin or non-fasting glucose

> 11mmol/L or fasting glucose >7mmol/L. Hypertension was defined as systolic blood pressure ≥140 mm Hg or diastolic pressure ≥90 mm Hg (mean of two measurements) or the use of antihypertensives. Alcohol consumption was categorized as less than 1 glass/day, 1-4 glasses/day for men and 1-2 glasses/day for women, and > 4 glasses/day for men and > 2 glasses/day for women. These were all assessed prospectively on a regular basis. Education was assessed according to the standard classification of education comparable to the international standard classification of education and was grouped into four categories "elementary education", "lower secondary education", "higher secondary education" and "tertiary education"

Statistical analysis

On the basis of previous study which found a prevalence of MI of 6.0% in AD patients vs. 2.4% controls and prevalences of class I and II obesity (BMI>35) 14.6% vs. 10.9% a sample size needed for a power of 80% and an error rate of 5% was 609 AD patients and 1217 non-AD subjects. Complete data analysis was performed for all variables excluding subjects missing the exposure or the outcome. Primary analyses were performed by binary logistic regression. In order to detect possible confounders and their effect on the AD/CHD association, two-way binary logistic regression was performed. Covariates with a $p<0.20$ effect on the AD-CHD association were added to the main models. In the main analyses three multivariable models were constructed and used to detect associations between AD (dependent variable) and the presence of CHD (independent variable). First model was adjusted for age and sex. Second model was additively adjusted for possible confounders from the two-way analysis and for cardiovascular risk factors differing across groups. The third model was adjusted for all known CVD risk factors mentioned under covariates. Sensitivity analysis was carried out on separate AD criteria in order to assess the contribution of the various components atopic dermatitis and to test how changes in definition of AD influenced the association. This makes the results more comparable to previous studies which used different definitions for AD. Also, AD being a diagnosis made by fulfilment criteria it is possible that a detected association is driven solely by one criterion and not the diagnosis of AD while a significant number of individuals are expected to fulfil some but not all necessary criteria, creating a category between exposed and unexposed. To reduce this contamination we also included an analysis in which the control consisted of subjects who explicitly never had been diagnosed with any form of eczema. Values of $p<0.05$ were considered significant. All data were analyzed using IBM SPSS 21.0 and R ver. 3.1.3, except for the sample size calculation; PS Power and Sample Size Calculations Version 3.0.

RESULTS

Patient characteristics

An FBSE was carried-out in a total of 4,386 participants but information concerning CHD and its risk factors was available for 4,342 (figure 1). Seven hundred twenty-three (16%) individuals were diagnosed with AD according to the criteria (table 1). These individuals reported having asthma, hay fever or house dust mite allergy more frequently than non-AD subjects (47% vs. 13%). Of these 723 subjects, 37% had a history of flexural involvement while only 5% had visible flexural dermatitis/eczema on examination (table 1) and 26% had any visible dermatitis. Hand eczema was the most prevalent form of visible eczema (hand eczema 13%, eczema on the extremities 12%, flexural eczema 5% and truncal eczema 4%). Of all AD cases 75% where already diagnosed with eczema by their family doctor or a dermatologist while this percentage was 13% in non-AD subjects.

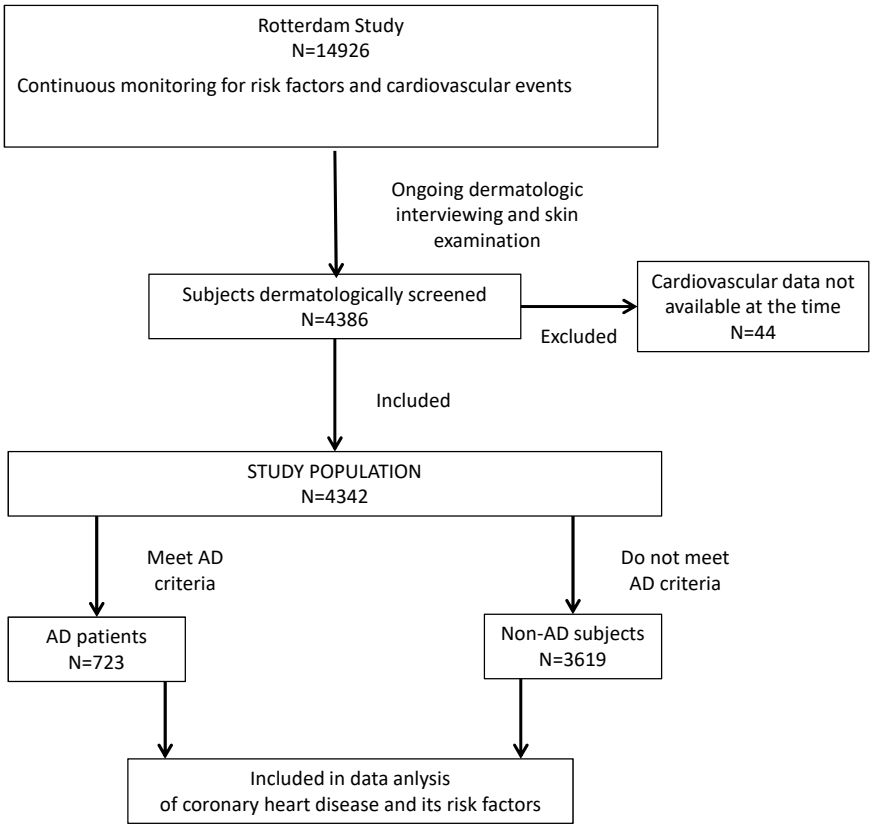

Figure 1. Flow diagram of the study population
A part of the Rotterdam Study population has been dermatologically screened and comprises the current study population

Table 1 Subject characteristics

	AD N=723	**No AD N=3619**
AD criteria		
Major criterion Itchy skin condition	723 (100%)	1331 (37%)
Minor criteria		
History of asthma	107 (15%)	148 (4%)
History of hay fever	215 (13%)	278 (8%)
History of HDM allergy	141 (20%)	192 (5%)
History of any of the above	339 (47%)	482 (13%)
History of involvement of skin creases	265 (37%)	246 (7%)
Generalized xerosis cutis on examination	640 (89%)	2352 (65%)
Visible dermatitis	198 (27%)	153 (4%)
Diagnosed with eczema by a physician	545 (75%)	471 (13%)
Other findings		
Physician diagnosed eczema before the age of 12	155 (21%)	121 (3%)
Visible dermatitis in creases	38 (5%)	17 (1%)
Visible hand dermatitis	96 (13%)	95 (3%)
BSA (mean)	0.52%	0.08%

Atopic dermatitis related findings in AD and non-AD subjects

AD and CVD risk factors

Atopic dermatitis patients were slightly younger (67.5 vs. 68.5 p=0.003) and less frequently male (37% vs 45% p<0.001). They also had slightly lower mean systolic blood pressure (142.3 vs 144.4)) and a higher mean HDL level (1.52 vs 1.48). No other statistically significant differences concerning the CVD risk factors were observed.

AD and CHD

There were 63 unique cases of CHD in the AD group and 276 in the non-AD group (8.7% vs. 7.6% p=0.358; table 2). The association between AD and CHD was significantly altered only by age, sex and HDL cholesterol in the bivariate analysis of confounders (table 3). A diagnosis of AD was not associated with CHD in the crude analysis OR 1.16; 95% CI (0.86-1.53) table 4.

After adjusting for sex and age in the first multivariate model a statistically significant association was found OR 1.42; 95% CI (1.05-1.89). This remained essentially unaltered in the second and third models with OR 1.42; 95% CI (1.04-1.90) and OR 1.54; 95% CI (1.09-2.15) respectively.

Table 2. Cardiovascular risk factors in AD patients and controls.

CHD risk factors	AD n=723	No AD n=3619	p-value
Age (years)	67.5 ± 8.6	68.5 ± 8.5	0.003
Sex (male)	266 (37%)	1552 (45%)	<0.001
Sys. blood pressure	142.3 ± 21.6	144.4 ± 22.0	0.038
Dias. blood pressure	83.6 ± 11.1	84.1 ± 11.2	0.219
Antihypertensive treatment	335 (46%)	1717 (46%)	0.684
Diuretics	112 (16%)	528 (15%)	0.571
BMI	27.7 ± 4.5	27.6 ± 4.4	0.475
Total cholesterol (mmol/L)	5.48 ± 1.10	5.47 ± 1.12	0.941
HDL (mmol/L)	1.52 ± 0.44	1.48 ± 0.43	0.013
Cholesterol lowering drugs	208 (29%)	1147 (30%)	0.444
Diabetes mellitus	106 (15%)	565 (15%)	0.918
Current smoking	97 (13%)	575 (16%)	0.105
Alcohol use	174 (24%)	868(24%)	1.000
CHD	63 (8.7%)	276 (7.6%)	0.358
Education			0.470
Elementary	79 (11%)	472 (13%)	
Lower secondary	285 (39%)	1411 (39%)	
Higher secondary	209 (29%)	1018 (28%)	
Tertiary	150 (21%)	718 (20%)	

Means and SD and percentages are reported where appropriate.
BMI, body mass index; HDL, high density lipoprotein; CHD, coronary heart disease

Table 3 Potential confounders

AD adjusted for	OR (95% CI) of CHD	p-value
No adjusting	1.16 0.86 1.53	0.320
Age	1.23 0.91 1.63	0.166
Sex	1.31 0.97 1.75	0.069
Current smoking	1.15 0.86 1.52	0.331
BMI	1.15 0.86 1.52	0.336
Total cholesterol	1.19 0.88 1.61	0.253
HDL cholesterol	1.25 0.93 1.65	0.138
Hypertension	1.19 0.88 1.58	0.240
Diabetes	1.16 0.86 1.54	0.311
Alcohol use	1.16 0.86 1.53	0.321
Education level	1.17 0.87 1.54	0.294
Use of antihypertensives	1.15 0.85 1.53	0.364
Use of diuretics	1.17 0.87 1.54	0.295

Left column contains potential confounders of the AD-CHD association and the middle the effects expressed as odds ratio

Sensitivity and subgroup analysis

In a sensitivity analysis we investigated the contribution of the individual AD criteria to the AD-CHD association. Of the individual criteria for AD only generally dry skin was significantly associated with CHD with adjusted ORs of 1.31; 95% CI (1.01-1.71) for model , OR 1.31; 95% CI (1.01-1.72) for model 2 and OR 1.34; 95% CI (1.00-1.81) for the fully adjusted model (table 4).

Table 4. The association between AD, components of AD diagnosis and CHD.

Variable	Crude OR (95% CI)	Model 1 OR (95% CI)	Model 2 OR (95% CI)	Model 3 OR (95% CI)
Diagnosis of AD	1.16 (0.86-1.53)	**1.42 (1.05-1.89)**	**1.42 (1.04-1.90)**	**1.54 (1.09-2.15)**
AD criteria tested separately				
Itch	1.02 (0.81-1.27)	1.14 (0.91-1.43)	1.13 (0.90-1.43)	1.19 (0.92-1.54)
Involvement of skin creases	0.80 (0.54-1.14)	0.97 (0.65-1.40)	0.97 (0.65-1.41)	0.99 (0.64-1.50)
Asthma, hay fever or house dust mite allergy	0.88 (0.65-1.17)	1.17 (0.86-1.57)	1.18 (0.86-1.60)	1.20 (0.84-1.68)
Generally dry skin	**1.40 (1.09-1.82)**	**1.31 (1.01-1.71)**	**1.31 (1.01-1.72)**	**1.34 (1.00-1.81)**
Visible dermatitis	0.82 (0.51-1.24)	0.80 (0.50-1.24)	0.83 (0.51-1.28)	0.98 (0.58-1.58)
Diagnosis of eczema by physician	0.99 (0.88-1.09)	1.00 (0.89-1.10)	0.99 (0.88-1.09)	1.04 (0.91-1.16)
Diagnosis of eczema before age of 12	0.95 (0.59-1.36)	1.20 (0.74-1.72)	1.27 (0.78-1.86)	1.34 (0.78-2.07)
Diagnosis of AD with Previous eczema diagnosis excluded from controls N=3326	1.46 (0.85-2.36)	**1.75 (1.00-2.90)**	**1.86 (1.06-3.10)**	1.80 (0.96-3.25)

Model 1 adjusted for sex and age;
Model 2 adjusted for age, sex and HDL; (continued)
Model 3 same as model 2 with diastolic blood pressure, use of antihypertensives, BMI, total cholesterol, cholesterol lowering drugs, diabetes mellitus, alcohol use, smoking and education level

Exclusion of individuals who were ever diagnosed with any eczema from the control group increased the ORs but was only statistically significant in model 1 and 2 OR 1.75; 95% CI (1.00-2.90) and OR 1.86; 95% CI (1.06-3.10).

DISCUSSION

The findings in our study suggest an association between CHD and AD which is independent of the traditional CVD risk factors such as smoking, obesity, high blood pressure and hypercholesterolemia with OR 1.42 95%CI [1.05-1.89]. Our findings are in concordance with the effect size found in a recent large prospective German study (aRR 1.32 vs. aOR 1.42).[10] In contrast to the studies from the U.S. and Asia we found no association between AD and traditional CVD risk factors.[9,18] This can be explained by the method of measuring of the risk factors and the use of a European study population. In previous studies the exposures were patient-reported while our study presents data obtained through validated methods. Supporting this notion is that in the same cohort of NHANES (National Health and Nutrition Examination Study) an inverse association between specific serum IgE and MI was found by Jaramillo et al. while Silverberg et al reported higher odds of having CVD and congestive heart failure in patients reporting flexural eczema. [6,19] This however, may also suggest that the increased risk of CVD in AD patients is in fact atopy independent and not driven by systemic inflammation. The scientific community is currently divided according to two main hypotheses explaining the associations found between CVD and AD. The first hypothesis is based on confounding by traditional CVD risk factors promoted by yet another study by Silverberg et al which in addition proposes that the increased risk of CVD in AD patients is attributable to a sedentary life style and sleep disturbances due to nightly itching. The second hypothesis is inflammation dependent, based on animal models, and states that AD leads to systemic inflammation and therefore enhanced atherosclerosis. [3,5,20,21] We were not able to assess the physical activity in our subjects but itch per se seems not to be associated with CHD in our study population. A more recent, well conducted study has demonstrated that adult AD patients in the U.S. lead a more sedentary life style than controls which may explain the increased risk of CVD.[22] We were however unable to reproduce the results from the high impact study by Silverberg et al when using the same definition of AD (patient reported physician's diagnosis).[5] In support, no positive association was found in underlying molecular pathways, between AD and CVD in terms of genetic risk variants and metabolomics in the aforementioned German study of multiple large prospective cohorts.[10]

On the other hand there is recent evidence from mouse models that acute disruption of the epidermal barrier by tape stripping or chronic disruption by aging lead to an increase in serum cytokines (IL-1α, IL-1β, IL-6, TNF-α) and amyloid A and that this systemic inflammation is reversible upon restoration of the barrier with topical petrolatum.[23] The finding that general xerosis cutis associates most significantly with CHD in our cohort

may have a basis in the same mechanisms and may be the pathophysiological bridge between AD and CVD.

The RS has been designed as a population-based cohort with the main objective being the epidemiological investigation of CVD minimizing residual confounding while the ascertainment of the outcome is standardized and limits misclassification bias. The fact that we analyzed hard outcome data on CHD rather than subclinical measures guarantees that there is no effect of increased detection of outcome from AD-status. Furthermore, the AD patients underwent the same examinations as the reference population excluding selection and detection bias while the diagnosis of AD has been made in accordance with validated UK working party's criteria. This is also the first study on AD and CHD where AD has been defined with a validated diagnostic tool. As these criteria were adapted some misclassification is possible. It would however lead to a dilution of the effect observed favoring the null hypothesis. Dowlatshahi and colleagues found an association between psoriasis vulgaris and CVD risk factors such as obesity, smoking and hypertension in this very same cohort.[24] This is in concordance with the data available from the literature and adds validity to our methods. The group that met the AD criteria comprised 16 % of the total study population. This is comparable to the prevalence of AD in children from the 40's and 50's (childhood period in our study population), as described in literature.[25] This is a cross-sectional study on prevalence of CHD, as such it carries with it some limitations and no causal relation can be inferred from the associations found. The age of onset of AD being a disease of childhood is much younger than the onset of CVDs. This makes it hard to study the association within a prospective cohort. The RS population is older than 55 years and may not represent the average AD patient, especially those with severe disease or systemically treated and hospitalized patients. In addition the findings are not generalizable to individuals younger than 55 years of age, a group which has been demonstrated to carry the greatest excess CVD risk in psoriasis. Moreover, there is a possibility of survival bias in this elderly population. Furthermore, the duration of AD exposure could not be measured and the increased risk of CHD can be argued to be present only during flare ups of this episodic skin condition. No data on flare ups and AD treatment were available but could have made possible further stratification of AD patients.

The clinical significance of our findings is not fully clear. Nevertheless, AD does not seem to be a good independent predictor of CHD as the crude analysis showed non-significant findings. Research into comorbidities should eventually lead to preventative measures but current CVD prevention programs revolve around curbing of the traditional risk factors.[26] It seems, from our and previous studies that the traditional risk factors do not seem to play a role in the AD-CVD association. Therefore, standard ways of screening will

not suffice a priori. What treatment should eventually be prescribed to AD patients at risk of CVD is one more scientific step further away. Measures of clinical relevance such as excess risk and number needed to screen should also be central in order to assess the clinical relevance of the study findings.

In conclusion, there seems to be an association between CHD and AD but this is not explained by traditional CVD risk factors. When considering all data available from current research there is clear inconsistency in findings. This indicates that the AD-CHD association may be a spurious one or at best a phenomenon of specific subgroups. Future studies should focus on prospective methodologies taking into account disease flare-ups and severity, the treatment, physical activity and sleep disturbances. Increased risk of CVD in AD is probably attributable to subgroups of more severe AD patients.

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REFERENCES

1. Williams H, Flohr C. How epidemiology has challenged 3 prevailing concepts about atopic dermatitis. J Allergy Clin Immunol 2006;118:209-13.
2. Bieber T. Atopic dermatitis. N Engl J Med 2008;358:1483-94.
3. Brunner PM, Silverberg JI, Guttman-Yassky E, et al. Increasing Comorbidities Suggest that Atopic Dermatitis Is a Systemic Disorder. J Invest Dermatol 2017;137:18-25.
4. Dhana K, Ikram MA, Hofman A, Franco OH, Kavousi M. Anthropometric measures in cardiovascular disease prediction: comparison of laboratory-based versus non-laboratory-based model. Heart 2015;101:377-83.
5. Silverberg JI, Greenland P. Eczema and cardiovascular risk factors in 2 US adult population studies. J Allergy Clin Immunol 2015;135:721-8 e6.
6. Silverberg JI. Association between adult atopic dermatitis, cardiovascular disease, and increased heart attacks in three population-based studies. Allergy 2015;70:1300-8.
7. Drucker AM, Li WQ, Cho E, et al. Atopic dermatitis is not independently associated with nonfatal myocardial infarction or stroke among US women. Allergy 2016;71:1496-500.
8. Su VY, Chen TJ, Yeh CM, et al. Atopic dermatitis and risk of ischemic stroke: a nationwide population-based study. Ann Med 2014;46:84-9.
9. Zhang A, Silverberg JI. Association of atopic dermatitis with being overweight and obese: a systematic review and metaanalysis. J Am Acad Dermatol 2015;72:606-16 e4.
10. Standl M, Tesch F, Baurecht H, et al. Association of atopic dermatitis with cardiovascular risk factors and diseases. J Invest Dermatol 2016.
11. Andersen YM, Egeberg A, Gislason GH, Hansen PR, Skov L, Thyssen JP. Risk of myocardial infarction, ischemic stroke, and cardiovascular death in patients with atopic dermatitis. J Allergy Clin Immunol 2016;138:310-2 e3.
12. Hofman A, Darwish Murad S, van Duijn CM, et al. The Rotterdam Study: 2014 objectives and design update. Eur J Epidemiol 2013;28:889-926.
13. Jacob SE, Goldenberg A, Nedorost S, Thyssen JP, Fonacier L, Spiewak R. Flexural eczema versus atopic dermatitis. Dermatitis 2015;26:109-15.
14. Hajdarbegovic E, Blom H, Verkouteren JA, Hofman A, Hollestein LM, Nijsten T. Atopic dermatitis is not associated with actinic keratosis: cross-sectional results from the Rotterdam study. Br J Dermatol 2016;175:89-94.
15. Jacquet A. Innate immune responses in house dust mite allergy. ISRN Allergy 2013;2013:735031.
16. Zeppa L, Bellini V, Lisi P. Atopic dermatitis in adults. Dermatitis 2011;22:40-6.
17. Leening MJ, Kavousi M, Heeringa J, et al. Methods of data collection and definitions of cardiac outcomes in the Rotterdam Study. Eur J Epidemiol 2012;27:173-85.
18. Yu SH, Silverberg JI. Association between Atopic Dermatitis and Depression in US Adults. J Invest Dermatol 2015;135:3183-6.
19. Jaramillo R, Cohn RD, Crockett PW, Gowdy KM, Zeldin DC, Fessler MB. Relation between objective measures of atopy and myocardial infarction in the United States. J Allergy Clin Immunol 2013;131:405-11 e1-11.
20. Yamanaka K, Nakanishi T, Saito H, et al. Persistent release of IL-1s from skin is associated with systemic cardio-vascular disease, emaciation and systemic amyloidosis: the potential of anti-IL-1 therapy for systemic inflammatory diseases. PLoS One 2014;9:e104479.
21. Halmerbauer G, Frischer T, Koller DY. Monitoring of disease activity by measurement of inflammatory markers in atopic dermatitis in childhood. Allergy 1997;52:765-9.

22. Silverberg JI, Song J, Pinto D, et al. Atopic Dermatitis Is Associated with Less Physical Activity in US Adults. J Invest Dermatol 2016;136:1714-6.
23. Hu L, Mauro TM, Dang E, et al. Epidermal Dysfunction Leads to an Age-Associated Increase in Levels of Serum Inflammatory Cytokines. J Invest Dermatol 2017.
24. Dowlatshahi EA, Kavousi M, Nijsten T, et al. Psoriasis is not associated with atherosclerosis and incident cardiovascular events: the Rotterdam Study. J Invest Dermatol 2013;133:2347-54.
25. Taylor B, Wadsworth J, Wadsworth M, Peckham C. Changes in the reported prevalence of childhood eczema since the 1939-45 war. Lancet 1984;2:1255-7.
26. Standaard Cardiovasculair Risicomanagement. NHG, 2015. (Accessed Third of January, 2017, at https://www.nhg.org/standaarden/volledig/cardiovasculair-risicomanagement.)

Chapter 5

General discussion

ATOPY

The best definition of atopy for clinical purposes is *a predisposition towards the development of atopic asthma, hay fever, food allergies and atopic dermatitis (AD)*. Therefore atopy is a spectral diathesis and an individual with any of the aforementioned conditions can be considered to be atopic to a certain degree. The best biomarker for general atopy is the specific serum IgE.[1,2] However, increased specific IgE levels are not always found in all asthma and AD patients as aspecific triggers also play a role.[1,3] Hence, in these disorders clinical criteria are needed to diagnose atopy. The most clinically and scientifically useful criteria to diagnose AD are the UK working Party's Criteria.[4] The pathogenesis of atopic disorders is most likely initiated by a skin barrier defect which is compensated for by the adaptive immune system leading to an activation of the T-helper lymphocyte 2 axis (Th-2 axis).[5] The systemic spread of this Th-2 polarization has been well documented and manifests itself as the atopic march and leads to the development of food allergies, hay fever and asthma.[6-8] The Th-2 mode of operating of the immune system is normally directed towards antigen (mainly macro-parasites) and toxin expulsion as well as barrier maintenance. The activation of this system in the absence of harmful antigens is called atopy. The overactivation of the Th-2 system in atopy is unique among immunologic disorders as it manifests itself in terms of behavior as sneezing (hay fever), coughing (asthma), vomiting (food allergies) and scratching (AD). Atopy-induced physical changes in the human body are hypertrophy of the mucosa and the skin and an increased secretion of mucus and tears. Given the barrier-based basis of atopy, it is not surprising that all components of the atopic diathesis take place at the border between the body and the external world, i.e. the mucosal and skin surfaces. Atopic patients are thus individuals in whom the barrier-based defenses have failed and in whom the hierarchically higher innate and adaptive immune responses have been overactivated.[9] As such, they are potentially at risk for other diseases, i.e. comorbidities, in which these parts of bodily defenses play a role. Next we will explain how atopy may predispose or even protect against other diseases as hypothesized at the beginning of this project.

Multiple SNPs (single nucleotide polymorphisms) have been shown to contribute to the development of overt clinical disease in the pathophysiological chain of atopy (FLG, IL4, IL4RA, SPINK5, CMA1, IL13, RANTES, CD14, DEFB1, GSTP1, IL18, NOD1 and TIM1).[10] The barrier-associated genes and genes associated with leukocyte and cytokine functions are most frequently affected.[11-13] The diminished barrier function invariably predisposes to a heavier exposure to toxins as well as to physical and infectious agents. At the same time the general and systemic polarization of the immune system towards a more Th-2-like profile affects the normal functioning of the immune system. Traditionally this is attributed to the Th-1 inhibiting effects of the Th-2 polarized immune system (see

introduction).[14] Therefore one of our hypotheses was that the impaired skin barrier of AD may predispose to UV-induced skin cancers and actinic keratosis (AK). In addition, we speculated that the Th-2 polarized immune system of atopic patients likely impairs the responses directed against tumors through the antagonism of the Th-1 axis, which also could lead to more skin cancer and AKs in AD patients (figure 1). The basis for this hypothesis was the idea that the general coordination of anti-tumor activity by the immune system is fully executed by the Th-1 axis. In line, the Th-1 cytokines IL-2 and interferon are the only cytokines efficacious in, and registered for the treatment of cancers.[15] However, IL-2 has pleiotropic effects beyond the domain of the Th-1 axis, illustrated by the fact that this cytokine stimulates B-cell proliferation and is even necessary for the proliferation and differentiation of Th-2 lymphocytes. These findings indicate the existence of more sophisticated effector functions of the Th-axes than explained by the traditional Th-1/Th-2 paradigm. [16,17] Moreover, the positive effect of IL-2 on the metastasized melanoma seems to be conveyed by a Th-2 cytokine, namely IL-24.[18,19] In addition to this apparent immunologic paradox, the association of AD with melanoma, keratinocyte cancer and actinic keratosis had not been tested in a population-based sample. We therefore set out to account for these shortcomings. The Th-1/Th-2 paradigm, and

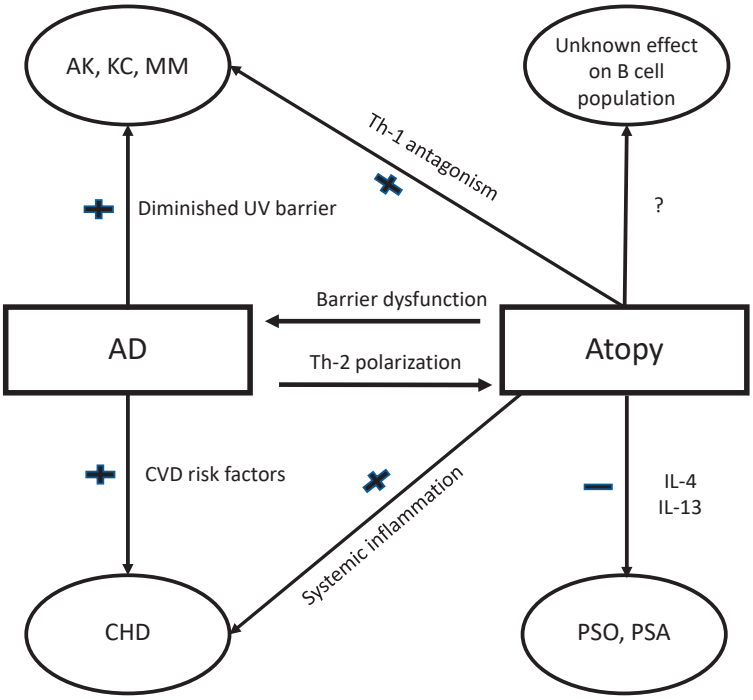

Figure 1. The hypotheses and their presumed mechanisms at the outset of the project
AD, atopic dermatitis; AK, actinic keratosis; CHD, coronary heart disease; CVD, cardiovascular disease; KC, keratinocyte cancer; MM, melanoma

more specifically the antagonism between Th-1 and Th-2 cells, have also been implied in the inverse associations found between atopy and inflammatory disorders, which are traditionally categorized as Th-1 driven diseases.[14] New discoveries in the past decade have shown a far more intricate adaptive immunological network with new Th-axes such as the T-regs, Th-9, Th-17 and Th-22, casting doubt on the universal applicability of the Th-1/Th-2 model. [20-24] Although the antagonism of the Th-1 and Th-17 systems by atopy has been demonstrated in rheumatoid arthritis and cutaneous psoriasis on tissue level, it has not been investigated in large patient cohorts of psoriasis vulgaris (PSO) and psoriatic arthritis (PSA).[14,25] Therefore, one of the objectives was to explore how atopy may affect the closely related autoinflammatory disorders of PSO and PSA, because both diseases are traditionally regarded as Th-1 and Th-17 mediated disorders (figure 1).[26,27] Another focus of this research was the effect of the Th-2 polarization on B-lymphocytes. Th-2 cells are known to induce isotype switching towards IgE producing in B-cells.[28,29] However the effects of atopy on the numbers of peripheral B-cells are unknown (figure 1).

Besides the associations between atopy with cancer and inflammatory disorders, there is increasing evidence linking atopy, more specifically AD, to systemic inflammation and cardiovascular disease (CVD).[14,30] The association between CVD and AD is currently explained by either the presence of traditional CVD risk factors in the AD patient or by the effects of systemic inflammation caused by AD. However, the evidence supporting the association between CVD and AD is poor. Previous research presented with several caveats such as the use of self-reported AD, CVD risk factors, and hospitalized AD patients, while CVD caused by systemic inflammation has not been robustly demonstrated in humans. [31,32] The main objective of this thesis was to explore the effects of atopy on the prevalence and severity of selected non-atopic diseases in the domains of oncology, immunology and cardiovascular disease.

FINDINGS

Atopy and inflammatory disorders

In the first part of chapter 2 the focus was on atopy and PSO. Although the pathogenesis of PSO shows multiple defects in the epidermal barrier formation, much like in AD, its occurrence shows no association with asthma, hay fever, or sensitization in our study.[33-35] We therefore conclude that atopic disorders are not a likely comorbidity of PSO, nor do they precede the development of PSO. Another conclusion that we came to is that atopy does not protect from the acquisition of PSO either. However, the results in patients with PSA were different, as we found a lower prevalence of atopy in this population. This

possibly indicates a protective effect of atopy on PSA through immunologic antagonism between atopy, which is a Th-2 dominated inflammatory response, and PSA, which in turn is a Th-1 and Th-17 skewed disease (figure 2). Because clinical atopy occurs much earlier in life than PSA, a reversed causality is not likely. Though in case of serological atopy (i.e. increased specific IgE), suppression of IgE secretion later in life in PSA patients cannot be ruled out. Despite the fact that PSO is also Th-1 and Th-17 driven, we found no inverse association of PSO with asthma, hay fever and sensitization. On the level of skin, the antagonism between AD and PSO has been well demonstrated however.[25] It is possible that in PSA patients, immunologic abnormalities are more systemically spread and play a greater role than the more localized cutaneous inflammation in PSO.[36] It is known that in PSA the expression of IL-17 is most dominant, while in PSO IL-22 is the most abundant and most active cytokine on tissue level. [37] This would make the antagonism by the Th-2 axis at a systemic level possibly more relevant in PSA compared to PSO. Polymorphisms in the interleukin-4 (IL-4) gene probably play a role in this phenomenon as this Th-2 cytokine is the main antagonist of theTh-1 and Th-17 axes.[38,39] Underscoring this theory, IL-4 has shown therapeutic value in clinical trials of PSO which was produced by IL-23 and Th-17 selective antagonizing effects.[40-42] In a genetic study, more severe, erosive PSA was associated with the IL-4 receptor 150V single nucleotide polymorphism (SNP).[43] Unfortunately, as pharmaceutical development of a non-patentable agent is not economically favorable, the effects of IL-4 on PSA have not yet been studied in clinical trials.

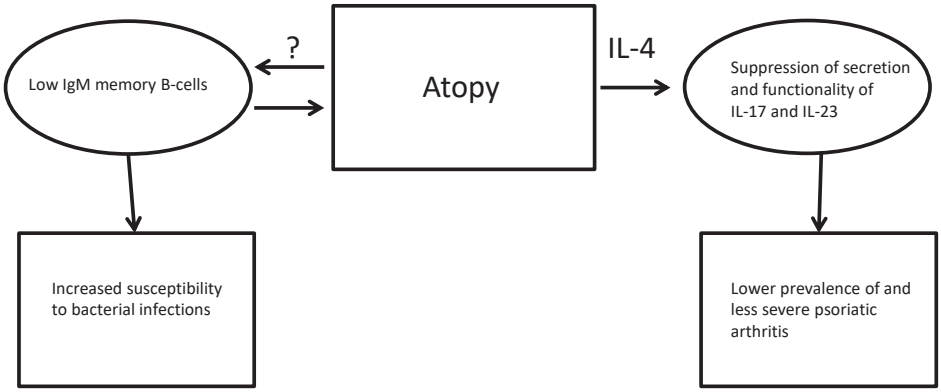

Figure 2. Atopy and the immune system, insights from this thesis and possible pathways

The second part of this chapter focused on the pathophysiology of AD and the role of the B-cells therein. AD pathophysiology can be appreciated as initially an immune deficiency of the innate immune system (epidermal barrier dysfunction), which is followed by changes in the adaptive immune system. However, not all cells of the adaptive

immune system are fully mapped in AD. The importance of the B-cells in AD has not been investigated with nearly half the intensity as the role of the T-lymphocytes. This is surprising as B-cells produce IgE, which is the hallmark protein and a biomarker of atopy. The IgE antibodies of AD patients have lower numbers of somatic mutations and antigen selection seems to play a less crucial role than in asthma and hayfever patients.[44] Still, most types of B-cells are relatively increased in the peripheral blood of AD patients. There is a systemic expansion of transitional and chronically activated CD27+ memory, plasmablasts, and IgE-expressing memory B-cell subsets and there is aberrant immunoglobulin class switching.[45,46] Previous research has also shown increased levels of CD27+ IgE memory cells with increased numbers of somatic hyper mutations.[47,48]

In chapter 2, part 2 we have demonstrated decreased absolute numbers of IgM-only memory B-cells in patients with AD. IgM-only memory B cells form the smallest compartment of B cells in the peripheral blood (about 10%) and selective IgM deficiency is associated with allergy and bacterial infections.[49] It is not clear whether decreased numbers of IgM memory B-cells play a role in the pathogenesis of AD or if they are an epiphenomenon resulting from constitutional immunological changes in atopy (figure 2). It is unlikely that this finding relates directly to Th-2 effects as IgM memory cells are T-cell independent.[50] Further exploration of B-cell abnormalities in AD patients is warranted with extensive AD phenotype specification and correlation with disease course details such as staphylococcal superinfections, eczema herpeticum and molluscum contagiosum infections.

Atopy and cancer

The controversial relationship between atopy and cancer has been the subject of many observational studies.[51] Most of these studies have focused on the relationship between asthma and internal malignancies, showing mostly inverse associations.[52,53] We have investigated the atopy-cancer link focusing on skin cancer and AD because both are primary cutaneous conditions. We hypothesized that epidermal barrier dysfunction and Th-1 suppression seen in AD skin could predispose to more UV-induced damage in the keratinocytes, subsequently leading to an increased risk of skin cancer and actinic keratosis (AK) in these patients.[54] Our findings disproved this hypothesis. Following this we concluded that increased surveillance for keratinocyte skin cancer in all AD patients is not warranted. Moreover, we found less severe AKs in community dwelling patients with AD. The case is obviously different for chronic severe AD patients who have been treated by UV-therapy and systemic immunosuppressants like ciclosporin A and azathioprine, which are known risk factors for AK and keratinocyte skin cancers.[55,56]

In the second study of this chapter focusing on melanoma and atopy, we found presumptive evidence of a protective effect of asthma against melanoma. There was a statistical power problem as only 184 melanoma patients could be recruited from our university hospital center, but several other cohort studies have corroborated our findings on this inverse association. [57-59] The low number of subjects was due to the fact that these patients, when diagnosed with melanoma, are mostly treated as soon as possible in non-academic hospitals and are therefore not referred to our institution. In conjunction, survivor bias could not be ruled out in this study. This may mean that the reduced prevalence of asthma we found in melanoma patients could be explained by the null hypothesis, if atopy were to confer a worse survival in melanoma.

The debate on atopy and cancer relationship has still not been settled however, which is probably due to the biologic heterogeneity of cancer. For example, melanoma and keratinocyte carcinomas are biologically different entities though both arise in the skin and both are called skin cancers. A second complicating factor in the studies is the considerable variation in how atopy is defined. Because of these issues, multiple mechanisms may be at play and the effect of atopy may therefore vary in different cancers. There are multiple, not mutually exclusive hypotheses explaining the antagonism between atopy and cancer (figure 3). Atopy seems to be especially protective for melanoma, pancreatic carcinoma, glioma and adult acute lymphoblastic leukemia according to a review from 2016.[60] Parts of the possible mechanism have also been revealed and can be divided into effects encountered within the innate and the adaptive immune systems. There are multiple components within the innate immune system which counteract carcinogenesis and are enhanced in atopy. Such innate immune mechanisms may therefore be responsible for the inverse association between atopy and cancer. The first hypothesis is only applicable for carcinomas i.e. epithelial malignancies. It states that, as IL-13 regulates the barrier repair by controlling keratinocyte maturation, its function is the clearance of tumor cells, toxins and carcinogens.[61] Knock-out models of IL-13 show an increased susceptibility to carcinogenesis due to an augmented cellular differentiation.[62,63] IL-13 is an important, well characterized and systemically increased (in atopy) cytokine belonging to the Th-2 axis. Thymic stromal lymphopoietin (TSLP) may enhance these tumor clearing effects of IL-13.[64] The second mechanism is the antagonism of the Th17 axis cytokines IL-6, IL-17, IL-21, IL-23 and TNF-α by the atopic Th-2 polarized immune system. These cytokines help build a tumor protective microenvironment and are key players in the propagation of carcinogenesis.[65] Lower levels of the Th-17 signature cytokines, as is the case in atopy, help to curb the tumor growth. Adding further evidence to the beneficiary effect of Th2 polarization is a recent study which found a positive correlation between serum eosinophils and median survival in advanced melanoma (35 vs. 16 months).[66] The reverse is true for the serum neutrophil numbers which are the Th-17

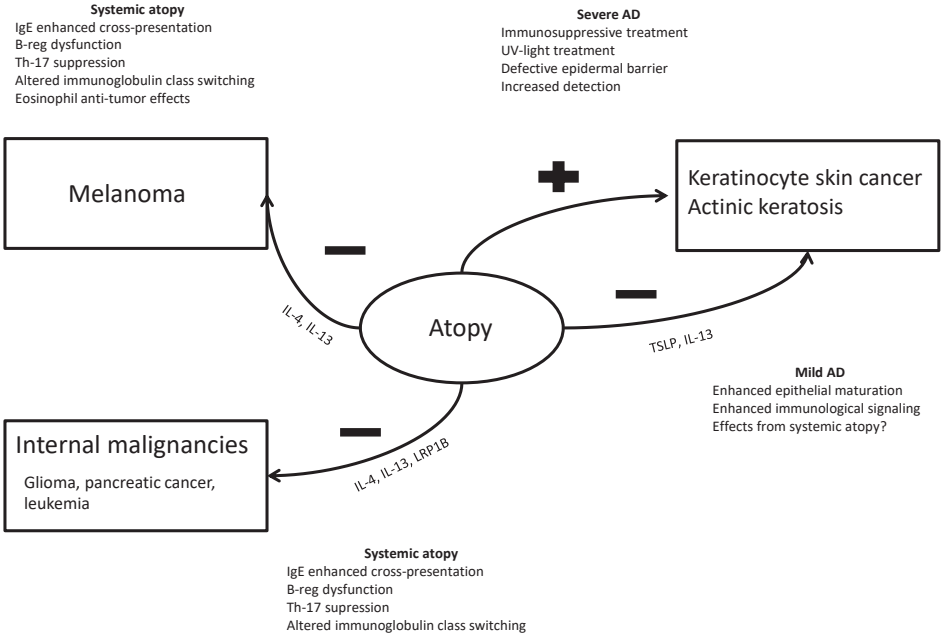

Figure 3. Links between atopy and cancer

innate immune system counterparts of the Th-2 driven eosinophils.[67] The eosinophils are not just bystanders but are proven to actively exert tumor rejecting activities such as guiding of CD8-positive T cells, macrophage polarization and tumor vasculature normalization.[68,69] On the adaptive side of the immune system multiple hypotheses have also been formed. SNPs of the low density lipoprotein related protein 1B (LRP1B) and interleukin-12 receptor A (IL12RA2) are associated with an 60% decreased risk of pancreatic cancer [70,71] LRP1B is involved in fat metabolism and antigen presentation. LRP1B-deficient macrophages are implied in progression of cancer while the aforementioned SNPs are also associated with asthma.[72,73] Several B-cell associated mechanisms exist in tumor immunology. B-cells are also key players in atopy and the adaptive immune system in general. The first aberration in of B-cells involved in these mechanisms is the attenuated activity of B-regulatory cells (B-regs) in atopy.[74] B-regs are known to infiltrate various tumors and diminish the activity of T-effector cells, natural killer cells and tumor-associated macrophages.[75] The dysfunctional B-regs in atopic individuals may be less tumor protective than in non-atopic individuals.[76] The second B-cell defect is the deviant class switching towards IgE instead of the classical progression to IgG4. It is currently known that tumor cells tend to shift the immune system in their microenvironment towards class switching to IgG4.[77] IgG4 is found in the microenvironment of

melanoma and is associated with worse prognosis.[78,79] Increased levels or functionality of IL-4 and IL-13 in atopic individuals prevent the development of this tumorigenic immunoglobulin by enhancing the class switching towards IgE.[28] An additional hypothesis involves yet another recently discovered function of the IgE molecule. Dendritic cells are able to cross-present (using MHC 1 instead of MHC 2) tumor antigens through the high affinity FC receptor of the IgE (FcƐRI). This leads to enhanced activation of cytotoxic T-cells and increased protection against tumors.[51,80] Interestingly, cross-presentation plays a major role in the immunity specifically against melanoma, pancreatic cancer and brain tumors.[81,82]

In the age of great advancements in immuno-oncology, research into the associations between atopy and cancer at any rate offers a better understanding of the immunologic mechanisms involved in carcinogenesis and tumor suppression. In best case, this knowledge may lead to enhancements of current immunologic therapies such as vaccines made to interact with FcƐRI receptor and induction of anti-tumor IgE antibodies.

Atopy and cardiovascular disease

The results of our study on the possible association between CHD and AD show that older, community dwelling patients with AD are more frequently affected by CHD than non-AD subjects. This relation seems to be independent of the traditional risk factors of CVD such as smoking, high BMI, diabetes mellitus and hypertension. This is in contrast to the previous studies performed in the United States and Asian countries, showing an association with traditional risk factors, but in agreement with the European studies.[30,83] Our results therefore indicate that if CVD is a direct comorbidity of AD it cannot be screened for by current methodologies (performed by the primary care physicians in the Netherlands) consisting of history taking and blood tests.[84] The main problem in current CVD-AD research is the selection of an appropriate, representative study population. Working with administrative data from non-medical databases and/or inclusion of hospitalized AD patients introduces misclassification and selection bias jeopardizing the external validity.[85] Hospitalized patients are more likely to be less adherent to both AD and CVD medication and they are more likely to undergo diagnostic procedures leading to detection bias. In our study, selection (indication) bias was minimized by the use of a community-based cohort in the Rotterdam study. In addition, known confounders were accounted for before data collection and during the analysis. One of the limitations in the ascertainment of atopic diseases is that the diagnosis of AD had been made during the adult life in all our subjects which allowed introduction of recall bias in the questionnaire based variables. Ideally, the diagnosis of atopy should be made during childhood as atopic disorders are then most prevalent. This helps to capture as many subjects as possible and avoids misclassification bias.

As to what explains the found association between AD and CHD, two theories are currently adhered to by the scientific community (figure 4). The proponents of the first theory argue that like in psoriasis, systemic inflammation of AD could accelerate atherosclerosis.[86] Although there is some evidence that AD may engender systemic inflammation in terms of increased serum levels of eosinophils, E-selectin and IgE, no study has explained the presumed mechanism by which systemic inflammation should lead to CVD.[87] Moreover, a direct comparison of genomic and metabolomic data of CVD and AD patients did not reveal any overlap.[88] The second theory is from a recent well conducted study which has shown that AD patients are more likely to live a sedentary life style and are less likely to engage in vigorous physical activity. Low physical activity is associated with an increased risk of CVD.[89] Sweat and exercise are known to exacerbate AD and induce itch and eczema affecting hands and feet; therefore patients with AD can be limited in their physical activities. In the same line of thinking, fatigue resulting from sleep disturbances may affect the incentive to exercise but could also independently increase the risk of CVD.[31,32] Behavioral adaptations in AD leading to an increased CVD risk should be explored further with the purpose of development of preventive programs. Currently the data on the subject of the AD-CVD association are too inconsistent which probably points out that a potential association does not stem from true pathophysiological mechanisms. Therefore, the contemporary evidence is insufficient to call for adjustments of clinical procedures or an increase in the surveillance for CVD in AD patients.

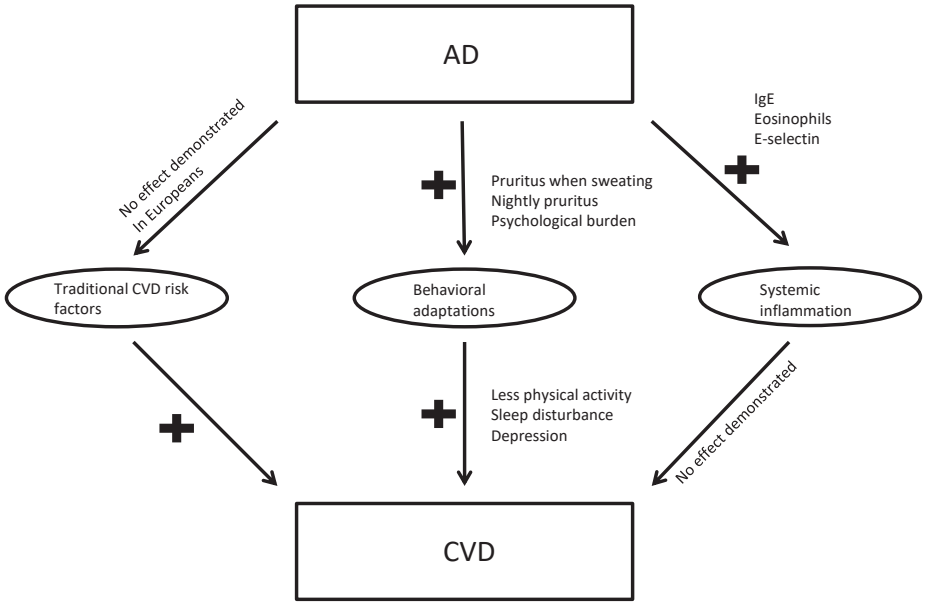

Figure 4. Putative theories explaining the association between CVD and AD.
AD, atopic dermatitis; CVD, cardiovascular disease

CONCLUSION: A NEW MODEL OF COMORBIDITY IN ATOPY

It is clear that our observations are based on various biologic mechanisms and that researchers should account for the different types of associations between diseases in future studies. When studying the co-occurrence of diseases one of the first questions which come into mind is whether the association found is based on causality or if it is a product of bias and chance. Discerning the two helps to understand the relevance of the research at hand. However, co-occurrence of diseases can be categorized in more than these two groups (figure 5). We have made a division between diathesis, direct comorbidity, indirect comorbidity and coexistence of diseases based on the mechanism of association. The distinction between these groups is very fluid and not strict as new research will move diseases from one category to another. An example for this phenomenon is eosinophilic esophagitis. While previously known to be associated with atopic disorders, eosinophilic esophagitis has just recently been added to the atopic diathesis group.[90] In a hypothetical example one could also imagine that a direct link could be found between fractures and atopic diseases mediated through osteoporosis due to vitamin-D deficiency.[91] Fractures would then belong to the direct comorbidity group. Because of the aforementioned fluidity, choosing the right topics for new research is impossible without regard of the previous scientific work. Of even more clinical importance and clinical relevance is the question whether the research into a particular comorbidity, be it direct or indirect, will lead to better outcomes or efficient disease prevention programs. The research in the field of psoriasis and cardiovascular comorbidities

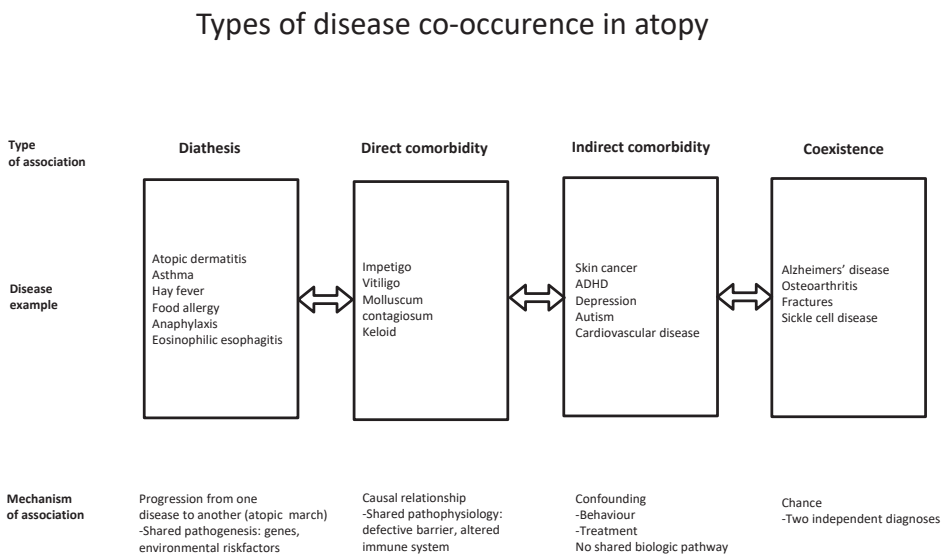

Figure 5. Categories of disease associations in atopy

has shown that vast quantities of high quality peer reviewed papers do not necessarily translate into a change of clinical practice.[85,92,93] Focus on the diseases from the atopic diathesis and direct comorbidity categories will likely have the highest yields in terms of advances in understanding of the pathogenetic mechanisms of atopy and implications for the daily clinical practice. Besides positive associations, atopy seems to be protective against Th-1 and Th-17 driven diseases. This is a phenomenon worth further investigation as this opens doors for a radically different concept in the treatment of immune disorders which is selective stimulation of the immune system instead of an immune system inhibition.

In conclusion, research into the comorbidities of atopy may be a very fruitful venture. Association does not automatically imply comorbidity. Atopy in itself is a systemic condition which by definition presents with multiple disorders making the patients and their treatment very complicated. Having additional comorbidities further increases the complexity of atopy cases. Fortunately, our research mainly provides evidence against an association with presumed comorbidities. We have shown that skin cancer is not a relevant comorbidity in AD. In addition atopy may actually confer protection against PSA and melanoma. The clinical relevance of the CHD-AD association needs further assessment but does not seem to necessitate changes in practice. Future research should be based on previous methodologically sound epidemiologic research in harmony with findings from plausible and thoroughly researched fundamental scientific work.

REFERENCES

1. Liu FT, Goodarzi H, Chen HY. IgE, mast cells, and eosinophils in atopic dermatitis. Clin Rev Allergy Immunol 2011;41:298-310.
2. Macglashan D, Jr. IgE and Fc{epsilon}RI regulation. Ann N Y Acad Sci 2005;1050:73-88.
3. Froidure A, Mouthuy J, Durham SR, Chanez P, Sibille Y, Pilette C. Asthma phenotypes and IgE responses. Eur Respir J 2016;47:304-19.
4. Andersen RM, Thyssen JP, Maibach HI. Qualitative vs. quantitative atopic dermatitis criteria - in historical and present perspectives. Journal of the European Academy of Dermatology and Venereology : JEADV 2016;30:604-18.
5. Boguniewicz M, Leung DY. Atopic dermatitis: a disease of altered skin barrier and immune dysregulation. Immunological reviews 2011;242:233-46.
6. Cork MJ, Danby SG, Vasilopoulos Y, et al. Epidermal barrier dysfunction in atopic dermatitis. J Invest Dermatol 2009;129:1892-908.
7. Dharmage SC, Lowe AJ, Matheson MC, Burgess JA, Allen KJ, Abramson MJ. Atopic dermatitis and the atopic march revisited. Allergy 2014;69:17-27.
8. Ker J, Hartert TV. The atopic march: what's the evidence? Annals of allergy, asthma & immunology : official publication of the American College of Allergy, Asthma, & Immunology 2009;103:282-9.

9. Spergel JM. From atopic dermatitis to asthma: the atopic march. Annals of allergy, asthma & immunology : official publication of the American College of Allergy, Asthma, & Immunology 2010;105:99-106; quiz 7-9, 17.
10. Barnes KC. An update on the genetics of atopic dermatitis: scratching the surface in 2009. J Allergy Clin Immunol 2010;125:16-29 e1-11; quiz 30-1.
11. Contopoulos-Ioannidis DG, Kouri IN, Ioannidis JP. Genetic predisposition to asthma and atopy. Respiration 2007;74:8-12.
12. Osmola A, Czarnecka-Operacz M, Silny W. Genetic aspects of atopy and atopic dermatitis. Acta Dermatovenerol Croat 2005;13:122-6.
13. Heinzmann A, Deichmann KA. Genes for atopy and asthma. Curr Opin Allergy Clin Immunol 2001;1:387-92.
14. Rabin RL, Levinson AI. The nexus between atopic disease and autoimmunity: a review of the epidemiological and mechanistic literature. Clinical and experimental immunology 2008;153:19-30.
15. Lee S, Margolin K. Cytokines in cancer immunotherapy. Cancers (Basel) 2011;3:3856-93.
16. Voss SD, Hank JA, Nobis CA, Fisch P, Sosman JA, Sondel PM. Serum levels of the low-affinity interleukin-2 receptor molecule (TAC) during IL-2 therapy reflect systemic lymphoid mass activation. Cancer Immunol Immunother 1989;29:261-9.
17. Sakaguchi S. Regulatory T cells: key controllers of immunologic self-tolerance. Cell 2000;101:455-8.
18. Jen EY, Poindexter NJ, Farnsworth ES, Grimm EA. IL-2 regulates the expression of the tumor suppressor IL-24 in melanoma cells. Melanoma Res 2012;22:19-29.
19. Poindexter NJ, Walch ET, Chada S, Grimm EA. Cytokine induction of interleukin-24 in human peripheral blood mononuclear cells. J Leukoc Biol 2005;78:745-52.
20. Sakaguchi S, Ono M, Setoguchi R, et al. Foxp3+ CD25+ CD4+ natural regulatory T cells in dominant self-tolerance and autoimmune disease. Immunological reviews 2006;212:8-27.
21. Harrington LE, Hatton RD, Mangan PR, et al. Interleukin 17-producing CD4+ effector T cells develop via a lineage distinct from the T helper type 1 and 2 lineages. Nat Immunol 2005;6:1123-32.
22. Dardalhon V, Collins M, Kuchroo VK. Physical attraction of Th9 cells is skin deep. Ann Transl Med 2015;3:74.
23. Jia L, Wu C. The biology and functions of Th22 cells. Adv Exp Med Biol 2014;841:209-30.
24. Veldhoen M, Hirota K, Westendorf AM, et al. The aryl hydrocarbon receptor links TH17-cell-mediated autoimmunity to environmental toxins. Nature 2008;453:106-9.
25. Eyerich S, Onken AT, Weidinger S, et al. Mutual antagonism of T cells causing psoriasis and atopic eczema. N Engl J Med 2011;365:231-8.
26. Raychaudhuri SP. Role of IL-17 in psoriasis and psoriatic arthritis. Clin Rev Allergy Immunol 2013;44:183-93.
27. Diani M, Altomare G, Reali E. T cell responses in psoriasis and psoriatic arthritis. Autoimmun Rev 2015;14:286-92.
28. Geha RS, Jabara HH, Brodeur SR. The regulation of immunoglobulin E class-switch recombination. Nat Rev Immunol 2003;3:721-32.
29. Akdis M, Akdis CA. IgE class switching and cellular memory. Nat Immunol 2012;13:312-4.
30. Zhang A, Silverberg JI. Association of atopic dermatitis with being overweight and obese: a systematic review and metaanalysis. J Am Acad Dermatol 2015;72:606-16 e4.
31. Silverberg JI, Greenland P. Eczema and cardiovascular risk factors in 2 US adult population studies. J Allergy Clin Immunol 2015;135:721-8 e6.

32. Silverberg JI. Association between adult atopic dermatitis, cardiovascular disease, and increased heart attacks in three population-based studies. Allergy 2015;70:1300-8.
33. Ye L, Lv C, Man G, Song S, Elias PM, Man MQ. Abnormal epidermal barrier recovery in uninvolved skin supports the notion of an epidermal pathogenesis of psoriasis. J Invest Dermatol 2014;134:2843-6.
34. Roberson ED, Bowcock AM. Psoriasis genetics: breaking the barrier. Trends in genetics : TIG 2010;26:415-23.
35. Guttman-Yassky E, Nograles KE, Krueger JG. Contrasting pathogenesis of atopic dermatitis and psoriasis--part I: clinical and pathologic concepts. J Allergy Clin Immunol 2011;127:1110-8.
36. Sakkas LI, Bogdanos DP. Are psoriasis and psoriatic arthritis the same disease? The IL-23/IL-17 axis data. Autoimmun Rev 2017;16:10-5.
37. Choy DF, Hsu DK, Seshasayee D, et al. Comparative transcriptomic analyses of atopic dermatitis and psoriasis reveal shared neutrophilic inflammation. J Allergy Clin Immunol 2012;130:1335-43 e5.
38. Ghoreschi K, Mrowietz U, Rocken M. A molecule solves psoriasis? Systemic therapies for psoriasis inducing interleukin 4 and Th2 responses. Journal of molecular medicine 2003;81:471-80.
39. Onderdijk AJ, Baerveldt EM, Kurek D, et al. IL-4 Downregulates IL-1beta and IL-6 and Induces GATA3 in Psoriatic Epidermal Cells: Route of Action of a Th2 Cytokine. J Immunol 2015;195:1744-52.
40. Guenova E, Skabytska Y, Hoetzenecker W, et al. IL-4 abrogates T(H)17 cell-mediated inflammation by selective silencing of IL-23 in antigen-presenting cells. Proceedings of the National Academy of Sciences of the United States of America 2015;112:2163-8.
41. Lotti T, Hercogova J. Successful treatment of psoriasis with low-dose per os interleukins 4, 10, and 11. Dermatologic therapy 2015;28:1-2.
42. Ghoreschi K, Thomas P, Breit S, et al. Interleukin-4 therapy of psoriasis induces Th2 responses and improves human autoimmune disease. Nature medicine 2003;9:40-6.
43. Rahman P, Snelgrove T, Peddle L, et al. A variant of the IL4 I50V single-nucleotide polymorphism is associated with erosive joint disease in psoriatic arthritis. Arthritis Rheum 2008;58:2207-8.
44. Kerzel S, Rogosch T, Struecker B, Maier RF, Kabesch M, Zemlin M. Unlike in Children with Allergic Asthma, IgE Transcripts from Preschool Children with Atopic Dermatitis Display Signs of Superantigen-Driven Activation. J Immunol 2016;196:4885-92.
45. Czarnowicki T, Gonzalez J, Bonifacio KM, et al. Diverse activation and differentiation of multiple B-cell subsets in patients with atopic dermatitis but not in patients with psoriasis. J Allergy Clin Immunol 2016;137:118-29 e5.
46. Oxelius VA. Imbalanced switch of the IGHG (immunoglobulin constant heavy G chain) Gm(bfn) genes in atopic childhood asthma. Allergy 2000;55:1063-8.
47. Berkowska MA, Heeringa JJ, Hajdarbegovic E, et al. Human IgE(+) B cells are derived from T cell-dependent and T cell-independent pathways. J Allergy Clin Immunol 2014;134:688-97 e6.
48. Berkowska MA, Driessen GJ, Bikos V, et al. Human memory B cells originate from three distinct germinal center-dependent and -independent maturation pathways. Blood 2011;118:2150-8.
49. Louis AG, Agrawal S, Gupta S. Analysis of subsets of B cells, Breg, CD4Treg and CD8Treg cells in adult patients with primary selective IgM deficiency. American journal of clinical and experimental immunology 2016;5:21-32.
50. Weller S, Braun MC, Tan BK, et al. Human blood IgM "memory" B cells are circulating splenic marginal zone B cells harboring a prediversified immunoglobulin repertoire. Blood 2004;104:3647-54.
51. Wulaningsih W, Holmberg L, Garmo H, et al. Investigating the association between allergen-specific immunoglobulin E, cancer risk and survival. Oncoimmunology 2016;5:e1154250.

52. Tennis P, Sherrill B, Fernandez C, Dolan C. Cancer risk in asthmatic populations. Annals of allergy, asthma & immunology : official publication of the American College of Allergy, Asthma, & Immunology 2005;95:354-60.
53. Cui Y, Hill AW. Atopy and Specific Cancer Sites: a Review of Epidemiological Studies. Clin Rev Allergy Immunol 2016;51:338-52.
54. Hajdarbegovic E, Verkouteren J, Balak D. Non-melanoma skin cancer: the hygiene hypothesis. Medical hypotheses 2012;79:872-4.
55. Maddox JS, Soltani K. Risk of nonmelanoma skin cancer with azathioprine use. Inflamm Bowel Dis 2008;14:1425-31.
56. Muellenhoff MW, Koo JY. Cyclosporine and skin cancer: an international dermatologic perspective over 25 years of experience. A comprehensive review and pursuit to define safe use of cyclosporine in dermatology. J Dermatolog Treat 2012;23:290-304.
57. Marasigan V, Morren MA, Lambert J, et al. Inverse Association Between Atopy and Melanoma: A Case-control Study. Acta Derm Venereol 2016.
58. Ji J, Shu X, Li X, Sundquist K, Sundquist J, Hemminki K. Cancer risk in hospitalised asthma patients. British journal of cancer 2009;100:829-33.
59. Kallen B, Gunnarskog J, Conradson TB. Cancer risk in asthmatic subjects selected from hospital discharge registry. Eur Respir J 1993;6:694-7.
60. Cui Y, Hill AW. Atopy and Specific Cancer Sites: a Review of Epidemiological Studies. Clin Rev Allergy Immunol 2016.
61. Palm NW, Rosenstein RK, Medzhitov R. Allergic host defences. Nature 2012;484:465-72.
62. Dalessandri T, Crawford G, Hayes M, Castro Seoane R, Strid J. IL-13 from intraepithelial lymphocytes regulates tissue homeostasis and protects against carcinogenesis in the skin. Nature communications 2016;7:12080.
63. Guinea-Viniegra J, Zenz R, Scheuch H, et al. Differentiation-induced skin cancer suppression by FOS, p53, and TACE/ADAM17. The Journal of clinical investigation 2012;122:2898-910.
64. Cipolat S, Hoste E, Natsuga K, Quist SR, Watt FM. Epidermal barrier defects link atopic dermatitis with altered skin cancer susceptibility. eLife 2014;3:e01888.
65. Monteleone G, Pallone F, Stolfi C. The dual role of inflammation in colon carcinogenesis. Int J Mol Sci 2012;13:11071-84.
66. Moreira A, Leisgang W, Schuler G, Heinzerling L. Eosinophilic count as a biomarker for prognosis of melanoma patients and its importance in the response to immunotherapy. Immunotherapy 2017;9:115-21.
67. Lino-Silva LS, Salcedo-Hernandez RA, Garcia-Perez L, Meneses-Garcia A, Zepeda-Najar C. Basal neutrophil-to-lymphocyte ratio is associated with overall survival in melanoma. Melanoma Res 2017.
68. Carretero R, Sektioglu IM, Garbi N, Salgado OC, Beckhove P, Hammerling GJ. Eosinophils orchestrate cancer rejection by normalizing tumor vessels and enhancing infiltration of CD8(+) T cells. Nat Immunol 2015;16:609-17.
69. Eftimie R, Bramson JL, Earn DJ. Modeling anti-tumor Th1 and Th2 immunity in the rejection of melanoma. J Theor Biol 2010;265:467-80.
70. Cotterchio M, Lowcock E, Bider-Canfield Z, et al. Association between Variants in Atopy-Related Immunologic Candidate Genes and Pancreatic Cancer Risk. PLoS One 2015;10:e0125273.
71. Li D, Duell EJ, Yu K, et al. Pathway analysis of genome-wide association study data highlights pancreatic development genes as susceptibility factors for pancreatic cancer. Carcinogenesis 2012;33:1384-90.

72. Gonias SL, Campana WM. LDL receptor-related protein-1: a regulator of inflammation in atherosclerosis, cancer, and injury to the nervous system. Am J Pathol 2014;184:18-27.
73. Li X, Howard TD, Zheng SL, et al. Genome-wide association study of asthma identifies RAD50-IL13 and HLA-DR/DQ regions. J Allergy Clin Immunol 2010;125:328-35 e11.
74. Braza F, Chesne J, Castagnet S, Magnan A, Brouard S. Regulatory functions of B cells in allergic diseases. Allergy 2014;69:1454-63.
75. Schwartz M, Zhang Y, Rosenblatt JD. B cell regulation of the anti-tumor response and role in carcinogenesis. J Immunother Cancer 2016;4:40.
76. van der Vlugt LE, Mlejnek E, Ozir-Fazalalikhan A, et al. CD24(hi)CD27(+) B cells from patients with allergic asthma have impaired regulatory activity in response to lipopolysaccharide. Clin Exp Allergy 2014;44:517-28.
77. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell 2011;144:646-74.
78. Karagiannis P, Villanova F, Josephs DH, et al. Elevated IgG4 in patient circulation is associated with the risk of disease progression in melanoma. Oncoimmunology 2015;4:e1032492.
79. Karagiannis P, Gilbert AE, Josephs DH, et al. IgG4 subclass antibodies impair antitumor immunity in melanoma. The Journal of clinical investigation 2013;123:1457-74.
80. Platzer B, Elpek KG, Cremasco V, et al. IgE/FcepsilonRI-Mediated Antigen Cross-Presentation by Dendritic Cells Enhances Anti-Tumor Immune Responses. Cell Rep 2015.
81. Huang AY, Golumbek P, Ahmadzadeh M, Jaffee E, Pardoll D, Levitsky H. Role of bone marrow-derived cells in presenting MHC class I-restricted tumor antigens. Science 1994;264:961-5.
82. Sigal LJ, Crotty S, Andino R, Rock KL. Cytotoxic T-cell immunity to virus-infected non-haematopoietic cells requires presentation of exogenous antigen. Nature 1999;398:77-80.
83. Kantor R, Kim A, Thyssen JP, Silverberg JI. Association of atopic dermatitis with smoking: A systematic review and meta-analysis. J Am Acad Dermatol 2016;75:1119-25 e1.
84. Standaard Cardiovasculair Risicomanagement. NHG, 2015. (Accessed Third of January, 2017, at https://www.nhg.org/standaarden/volledig/cardiovasculair-risicomanagement.)
85. Samarasekera EJ, Neilson JM, Warren RB, Parnham J, Smith CH. Incidence of cardiovascular disease in individuals with psoriasis: a systematic review and meta-analysis. J Invest Dermatol 2013;133:2340-6.
86. Brunner PM, Silverberg JI, Guttman-Yassky E, et al. Increasing Comorbidities Suggest that Atopic Dermatitis Is a Systemic Disorder. J Invest Dermatol 2017;137:18-25.
87. Yamashita N, Kaneko S, Kouro O, Furue M, Yamamoto S, Sakane T. Soluble E-selectin as a marker of disease activity in atopic dermatitis. J Allergy Clin Immunol 1997;99:410-6.
88. Standl M, Tesch F, Baurecht H, et al. Association of atopic dermatitis with cardiovascular risk factors and diseases. J Invest Dermatol 2016.
89. Silverberg JI, Song J, Pinto D, et al. Atopic dermatitis is associated with less physical activity in US adults. J Invest Dermatol 2016.
90. Furuta GT, Katzka DA. Eosinophilic Esophagitis. N Engl J Med 2015;373:1640-8.
91. Man L, Zhang Z, Zhang M, et al. Association between vitamin D deficiency and insufficiency and the risk of childhood asthma: evidence from a meta-analysis. Int J Clin Exp Med 2015;8:5699-706.
92. Maybury CM, Barker JN, Smith CH. Psoriasis and cardiovascular disease: where is the risk? J Invest Dermatol 2013;133:2308-11.
93. Stern RS. Psoriasis is not a useful independent risk factor for cardiovascular disease. J Invest Dermatol 2010;130:917-9.

Chapter 6

Summary
Samenvatting

ENGLISH SUMMARY

Chapter 1 is the general introduction to this thesis. We discuss the concepts of atopy and comorbidity and how these are applied in the clinical and scientific fields. Both have been subject to change over time. There is a brief overview of the four atopic disorders, atopic dermatitis, food allergy, asthma and hay fever. The pathophysiology is discussed in depth with an emphasis on the epidermal barrier dysfunction and Th2-axis activation. This is used as a basis for the explanation of the comorbidity model of atopic dermatitis. This chapter also gives an overview of previously reported comorbidities of AD and the level of evidence for these reports. It reveals the multitude of categories of and individual comorbidities of AD.

Chapter 2 contains the data from the studies in which have employed laboratory measurements. In the first part we report the findings of our cross-sectional cohort study where the life time prevalences of atopic disorders and IgE-sensitization are compared between psoriasis vulgaris patients, psoriatic arthritis patients and controls. From this study it is clear that neither psoriasis vulgaris nor psoriatic arthritis are comorbidities of atopy. Moreover, atopy is less frequently found in psoriatic arthritis patients. These patients were 4 times less frequently affected by asthma and were half times as much sensitized to common aeroallergens. In addition the ones who were sensitized showed attenuated phenotypes of their disease. Second part of this chapter shows that patients with atopic dermatitis have decreased absolute levels of IgM-only B-memory cells. It is not clear what this means in terms of disease course or comorbidity.

Chapter 3 focuses on the skin malignancies and premalignancies as a comorbidity of atopy. In the first part we explore the possible pathomechanisms and evidence for this association. There is ample evidence that the epidermal barrier together with breakdown products of filaggrin minister to the protection against UV-radiation. Therefore, damage to this barrier may lead to UV-induced skin cancer. However this is not applicable to community dwelling mild cases of AD as we have found in the second part of this chapter. There is no association between AD and actinic keratosis which are a prevalent premalignancie of the skin and a good marker of UV-induced DNA-damage. In the third part no positive association was found between AD and melanoma, the deadliest skin cancer. However our data suggest that asthma may confer protection against this malignancy.

Chapter 4 provides insight into the relationship between AD and cardiovascular disease (CVD). In this study nested within the Rotterdam Study cohort we have identified AD patients and compared them to controls in terms of CVD risk factors and coronary heart

disease prevalence. The Rotterdam Study has been specifically designed to address question concerning CVD. It contains high quality, validated and structurally verified data. We have found no association between AD and the traditional CVD risk factors. However AD was associated with coronary heart disease. Which seems to be an independent relation.

Chapter 5 is the general discussion of the studies performed for this thesis and the literature of studies by others performed before or after the publication of these papers. It provides scientific and clinical context to our findings. We illustrate the links between atopy, cancer, autoinflammatory/immune disorders and cardiovascular disease. At the end we demonstrate how comorbidity research could account for various methodological pitfalls and biases.

NEDERLANDSE SAMENVATTING

Hoofdstuk 1 is de algemene introductietekst van dit proefschrift. Hierin worden de concepten "atopie" en "co-morbiditeit" besproken en de wijze waarop deze begrippen toegepast worden in het wetenschappelijke en het klinische veld. Beide termen zijn onderhevig geweest aan veranderingen van de tijdsgeest. We geven verder een overzicht van de vier klassieke atopische aandoeningen: atopisch eczeem, voedingsallergie, astma en hooikoorts. De pathofysiologie van atopie wordt uitgebreid besproken aan de hand van het model van epidermale barrieredysfunctie en Th2-as activatie. Dit model wordt ook gebruikt om de mogelijke co-morbiditeiten van atopische aandoeningen te verklaren. Dit hoofdstuk maakt verder een opsomming van eerder co-morbiditeitsonderzoek en laat hier mee een veelvoud aan ziektebeelden zien.

Hoofdstuk 2 bevat de studies waarin de nadruk ligt op laboratoriumbepalingen. In het eerste gedeelte van dit hoofdstuk verslaan we de bevindingen van de cross-sectionele studie waarin we de prevalentie van atopische aandoeningen en IgE-sensibilisatie hebben vergelijken tussen controles en patiënten met psoriasis vulgaris en artritis psoriatica. Deze studie wijst uit dat beide ziektebeelden geen co-morbiditeit zijn van atopie. Sterker nog, atopie wordt minder frequent gezien in patienten met artritis psoriatica. Deze patienten hadden een viervoudig verlaagde kans op astma en tweevoudig verlaagde kans op sensibilisatie door aeroallergenen. De gesensibiliseerde patienten hadden zelfs milder verlopende artritis psoriatica. Het tweede deel laat zien dat patienten met atopisch eczeem verlaagde absolute aantallen IgM-only B-geheugencellen hebben. Het is niet duidelijk wat hiervan de betekenis is betreffende het ziektebeloop of co-morbiditeitenlast.

Hoofdstuk 3 focust op huid(pre)maligniteiten als co-morbiditeiten van atopie. In het eerste deel verkennen we de mogelijke pathomechanismen en het bewijs voor de associaties. Er is voldoende bewijs dat de epidermale barrière samen met de afbraakproducten van filaggrin meehelpen beschermen tegen Uv-straling. Wij concluderen dat schade aan deze barrière kan leiden tot Uv-geïnduceerde kanker. In deel twee blijkt dat dit mechanisme geen effect sorteert bij milde gevallen van atopisch eczeem binnen de gewone populatie. Er is geen associatie tussen actinische keratosen en atopisch eczeem. Actinische keratosen zijn een veelvoorkomende premaligniteit van de huid en een goede graadmeter van Uv-schade aan de huid. In het derde deel hebben we laten zien dat er geen positieve associatie is tussen atopisch eczeem en melanoom, de dodelijkste huidkanker. Niettemin lijkt het hebben van astma de kans op het melanoom te verlagen.

Hoofdstuk 4 verschaft inzicht in de relatie tussen atopisch eczeem en cardiovasculair lijden. In deze studie, onderdeel van de Rotterdam Study cohort hebben we patienten met atopisch eczeem geïdentificeerd en vergeleken met controles voor wat betreft cardiovasculaire risicofactoren en coronaire hartziekten. De Rotterdam Study werd specifiek ontworpen met cardiovasculaire aandoeningen voor de ogen. Het verstrekt data van hoge kwaliteit die regelmatig gevalideerd en geverifieerd wordt. We hebben geen relatie gevonden tussen het atopisch eczeem en de traditionele cardivasculaire risicofactoren. Er was echter wel een associatie tussen atopisch eczeem en coronaire hartziekten. Dit lijkt een onafhankelijke relatie te zijn.

Hoofdstuk 5 bevat de algemene discussie van de verrichte studies en van de literatuurstudie verricht voor dit proefschrift. Hierin pogen wij onze bevindingen in een bredere wetenschappelijke en klinische context te plaatsen. Mogelijke links tussen atopie en kanker, autoinflammatoire aandoeningen en cardiovasculaire aandoeningen worden besproken. Hier kaarten wij ook de methodologische valkuilen van co-morbiditeitsonderzoek aan.

Chapter 7

Appendices

ABBREVIATIONS

AD;	Atopic dermatitis
AK;	Actinic keratosis
AR;	Allergic rhinitis
BCC;	Basal cell carcinoma
CASPAR;	Classification Criteria for Psoriatic Arthritis
CHD;	Coronary heart disease
CI;	Confidence interval
CVD;	Cardiovascular disease
DAS 28;	Disease Activity Score 28
HAQDI;	Health assessment questionaire-disability index
HAQ-VAS;	Health assessment questionnaire visual analogue scale
IRR;	Incidence rate ratio
KC;	Keratinocyte cancer
OR;	odds ratio
PASI;	Psoriasis Area and Severity Index
PSO;	Psoriasis vulgaris or chronic plaque type psoriasis
PSA;	Psoriatic arthritis
RA;	Rheumatoid arthritis
RS;	Rotterdam study
SCC;	Squamous cell carcinoma
SD;	Standard deviation
Th;	T-helper cell
UVB;	Ultraviolet-B
UVR;	Ultraviolet radiation
95% CI;	95% confidence interval

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LIST OF PUBLICATIONS

1. Hajdarbegovic E, van der Leest RJT, Munte K, Thio HB, Neumann HAM. Neoplasms of the Facial Skin. *Clinics in plastic surgery* 2009; **36**(3): 319-34.
2. Hajdarbegovic E, Thio HB. Sensitive skin. *Nederlands Tijdschrift voor Dermatologie en Venereologie* 2010; **20**(7): 398-400.
3. Hajdarbegovic E, Van Der Leest RJT, Van Der Snoek EM, Thio HB. Herpes simplex lymphangitis. *Nederlands Tijdschrift voor Dermatologie en Venereologie* 2010; **20**(6): 354-5.
4. Hajdarbegovic E, Van Der Snoek EM, Ossewaarde JM, Van Der Meijden WI, Thio HB. Just a penile nodule. *Sexually transmitted diseases* 2010; **37**(4): 279-80.
5. Balak DMW, Hengstman GJD, Hajdarbegovic E, Van Den Brule RJP, Hupperts RMM, Thio HB. Impact on health-related quality of life by cutaneous adverse events associated with long-term disease-modifying therapy in multiple sclerosis: The derMiS study. *Multiple Sclerosis* 2012; **18**(4): 487.
6. Berkowska MA, Heeringa JJ, Hajdarbegovic E, et al. Small populations with major implications? identification of two IGE+ memory B-cell subsets in human blood-implications for primary immunodeficiencies. *Journal of Clinical Immunology* 2012; **32**: S259.
7. Hajdarbegovic E, Balak D. Metastatic inguinal lymphadenopathy. *New England Journal of Medicine* 2012; **366**(16): 1526.
8. Hajdarbegovic E, Balak DMW, Thio HB. The advantage of atopy. *Nederlands Tijdschrift voor Dermatologie en Venereologie* 2012; **22**(2): 109-12.
9. Hajdarbegovic E, Thio HB. Itch pathophysiology may differ among ethnic groups. *International journal of dermatology* 2012; **51**(7): 771-6.
10. Hajdarbegovic E, Thio HB. A neonate with a skin defect. *Nederlands tijdschrift voor geneeskunde* 2012; **156**(16).
11. Hajdarbegovic E, Verkouteren J, Balak D. Non-melanoma skin cancer: The hygiene hypothesis. *Medical hypotheses* 2012; **79**(6): 872-4.
12. Balak DMW, Hengstman GJD, Hajdarbegovic E, van den Brule RJP, Hupperts RMM, Thio HB. Prevalence of cutaneous adverse events associated with long-term disease-modifying therapy and their impact on health-related quality of life in patients with multiple sclerosis: A cross-sectional study. *BMC neurology* 2013; **13**.
13. Hajdarbegovic E, Nijsten T, Westgeest A, Habraken F, Hollestein L, Thio B. Decreased prevalence of atopic features in patients with psoriatic arthritis, but not in psoriasis vulgaris. *Journal of the American Academy of Dermatology* 2013; **68**(2): 270-7.
14. Berkowska MA, Heeringa JJ, Hajdarbegovic E, et al. Human IgE+ B cells are derived from T cell-dependent and T cell-independent pathways. *Journal of Allergy and Clinical Immunology* 2014; **134**(3): 688-97.e6.

15. Hajdarbegovic E, Atiq N, Van Der Leest R, Thio B, Nijsten T. Atopic dermatitis is not a protective factor for melanoma but asthma may be. *International journal of clinical oncology* 2014; **19**(4): 708-11.
16. Hajdarbegovic E, Kamphuis L, Van Laar J, Van Hagen M, Nijsten T, Thio B. Prevalence of atopic diseases in patients with sarcoidosis. *Allergy and Asthma Proceedings* 2014; **35**(4): e57-e61.
17. Hajdarbegovic E, Nijsten T. A man with a painful and swollen ankle. *Nederlands tijdschrift voor geneeskunde* 2014; **158**(11).
18. Hajdarbegovic E, Thio B, Nijsten T. Lower lifetime prevalence of atopy in rheumatoid arthritis. *Rheumatology international* 2014; **34**(6): 847-8.
19. Balak D, Hajdarbegovic E. PML in patients treated with dimethyl fumarate: To the editor. *New England Journal of Medicine* 2015; **373**(6): 582-3.
20. Hajdarbegovic E, Bloem A, Balak D, Thio B, Nijsten T. The association between atopic disorders and keloids: A case-control study. *Indian journal of dermatology* 2015; **60**(6): 635.
21. Van Boheemen S, Fouchier RAM, Hajdarbegovic E. In reply. *JAMA dermatology* 2015; **151**(4): 458.
22. Van Boheemen S, Jones T, Muhlemann B, Feltkamp MC, Fouchier RAM, Hajdarbegovic E. Cidofovir gel as treatment of follicular spicules in multiple myeloma. *JAMA dermatology* 2015; **151**(1): 82-4.
23. Balak D, Hajdarbegovic E. More on PML in Patients Treated with Dimethyl Fumarate. Author reply. *The New England journal of medicine* 2016; **374**(3): 295.
24. Hajdarbegovic E, Blom H, Verkouteren JAC, Hofman A, Hollestein LM, Nijsten T. Atopic dermatitis is not associated with actinic keratosis: Cross-sectional results from the Rotterdam study. *British Journal of Dermatology* 2016.
25. Heeringa JJ, Hajdarbegovic E, Thio HB, van Zelm MC. Systemic B-cell abnormalities in patients with atopic dermatitis? *Journal of Allergy and Clinical Immunology* 2016.
26. Hajdarbegovic E, Balak D. Percutaneous temporal artery ligature. *Journal of the American Academy of Dermatology 2016. Epub*
27. Balak D, Hajdarbegovic E. Nemolizumab in Atopic dermatitis. *The New England journal of medicine* 2017.
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Enes Hajdarbegovic was born on the first of january 1982 in Zvornik, Bosnia and Herzegovina to Rizah Hajdarbegovic and Hanumica Hajdarbegovic-Kapidzic. He has a brother, Dino who is an aerospace engineer and much better looking. In 1995 the four of them have escaped from Bosnia and Herzegovina (from the Balkan war) to the Netherlands. Enes currently works as a dermatologist in Rotterdam, Capelle aan den Ijssel and Goes. He lives with his wife, Anneke de Vos, who is a better physician and a better person in general. They have met during their studies. They have produced, and continue to mold, two daughters, Nora and Iza. After completing his studies at the Erasmus University Rotterdam, faculty of medicine, Enes has worked as an intern in various hospitals in the region. He started as a registrar in 2010 at the department of dermatology at the Erasmus Medical Centre where this PhD project was initiated. He plans to continue to improve his humanly, clinical and scientific skills.

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