

Soluble cytokine receptor CD30 in atopic disorders: a case-control study

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U. LATZA*, S. DAVIS*†, D. WILHELM‡, B. McKNIGHT§, M. SEYFARTH¶
and H. STEIN**

*Department of Epidemiology, University of Washington, Seattle, WA, USA, †Program in Epidemiology, Division of Public Health Sciences, Fred Hutchinson Cancer Research Center, University of Washington, Seattle, WA, USA, ‡proDERM Institute for Applied Dermatological Research, Schenefeld/Hamburg, Germany, §Department of Biostatistics, University of Washington, Seattle, WA, USA, ¶Institute for Immunology and Transfusion Medicine, Medical University of Lübeck, Germany and **Institute for Pathology, UKBF, Free University of Berlin, Germany.

Summary

Background The cytokine receptor CD30 is an activation marker of T cells which preferentially associates with the production of the TH2 cytokine IL-4. Therefore, it may potentially be a candidate marker for atopic disorders and a target molecule for new therapeutic approaches.

Objective To test the hypothesis that elevated levels of soluble CD30 (sCD30) are significantly associated with atopic disorders after adjustment for other predictors of atopy.

Methods The presence of elevated sCD30 (≥ 20 U/mL) in atopic disorders was evaluated in a nested case-control study. Cases ($n=60$) were blood donors with specific IgE antibodies, total serum levels of IgE ≥ 100 kU/L and presence or history of allergic symptoms. Controls ($n=59$) were blood donors without presence or history of allergic symptoms and serum levels of IgE < 50 kU/L. sCD30 was determined from serum samples by an enzyme-linked immunosorbent assay. Odds ratios (OR) and confidence intervals (CI) were calculated from logistic regression coefficients.

Results Mean sCD30 levels for cases were 75 U/mL (SD 110 U/mL) as compared with 35 U/mL (SD 59 U/mL) for controls. Serum levels of sCD30 were elevated in 65% of cases and 32% of controls (OR 3.9, 95% CI 1.8–8.4). The odds ratio for elevated sCD30 as a predictor of atopic disorders slightly decreased to 3.7 after controlling for smoking, age and gender. Blood eosinophilia which was a strong predictor of atopy (OR 11.7) was a weak confounder of the association between sCD30 levels and atopic disorders. Family history of allergy, another strong predictor of atopy (OR 8.6), did not confound the association.

Conclusions The results are consistent with the hypothesis that CD30 is involved in the pathogenesis of atopic disorders independent of eosinophilia and family history of allergy.

Keywords: case-control study, biomarker, CD30, atopy, cytokine receptor, TH2 cells, eosinophilia, IgE, family history of allergy

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Introduction

CD30 is a member of the tumour necrosis factor (TNF) receptor family [1] which can be shedded [2]. Interaction of

the receptor with the CD30 ligand [3], a TNF-like cytokine, may play a role in the co-stimulation of lymphatic cells under pathological and normal conditions. Soluble CD30 [4] (sCD30) has been demonstrated in the sera of viral [5–7], malignant [8–10], and autoimmune disorders [11–15]. In healthy individuals, CD30 is restricted to a subset of activated B and T lymphocytes in lymphoid tissues [16].

Correspondence: U. Latza, Institute for Social Medicine, Medical University of Luebeck, St. Juergen-Ring 66, D-23564 Luebeck, Germany.

The majority of CD30⁺ T cells are activated helper cells (CD4⁺, CD25⁺) of the memory subpopulation (CD45RO⁺) [16,17]. T-helper (TH) cells can be grouped according to their cytokine profile [18]. In this model, TH1 cells produce cytokines which strongly promote cell-mediated immune responses. TH2 cells secrete, among other cytokines, IL-4 and IL-5 which are involved in humoral immunity and allergic responses such as up-regulated IgE production and eosinophilia. Based on the finding that CD30 was preferentially expressed by TH2 cell clones [19], CD30 has been proposed as a marker of TH2 cells [20]. Recent data have demonstrated that CD30 is induced on activated T cells that respond to IL-4 [21]. Consequently, CD30 may potentially be a candidate marker for atopic disorders.

In a study by Del Prete and co-workers, CD30 expression was observed simultaneously with the production of IL-4 on blood lymphocytes of grass pollen-sensitive patients during the pollen season when stimulated *in vitro* with TH2 inducing antigens whereas CD30 was not detected when cells were stimulated with TH1-inducing antigens [19]. Further, sCD30 was found in cell culture supernatants from the T-cell clones of these atopic individuals [19].

So far, the presence of the cellular or soluble CD30 has never been tested in a controlled epidemiological study but only in case series. The authors evaluated sCD30 in a nested case-control study among blood donors. For this purpose, sCD30 serum levels of atopic individuals were compared with sCD30 levels of nonatopic controls. Further, the relationship of sCD30 with total serum levels of IgE (sIgE), eosinophil counts and family history of allergy was investigated.

Materials and methods

Description of the population for the nested case-control study

In a cross-sectional study, blood donors aged 18–65 years at their first visit to the University hospital in Lübeck, Germany in 1992 and 1993 ($n = 1251$) were invited to participate in a study on allergy prevalence [22]. Individuals who consented ($n = 1125$) were asked to fill out a questionnaire in which demographic information and data on symptoms and manifestations of airway diseases, asthma and other allergic disorders as well as first degree family history of allergy, and hypersensitivity reactions were obtained [22]. All participating blood donors were examined for physical signs for atopy. Blood samples were obtained for laboratory tests. Individuals with positive viral tests for the presence of hepatitis B virus surface antigen and antibodies against HIV, hepatitis C virus (HCV) and syphilis (*Treponema pallidum*) were excluded. Further exclusion criteria were cancer, autoimmune disease,

and the presence of a severe cold. Blood donors participating in the study were not taking hypertension drugs, aspirin or nonsteroidal anti-inflammatory drugs. After exclusion, the sample consisted of 1088 blood donors. The mean age was 31.4 ± 11 years for females (48.9%) and 31.5 ± 10.7 years for males.

Selection of cases and controls

A retrospective case-control study was conducted at the University of Washington, in 1996 which was nested within the cross-sectional study among blood donors [22]. The study was exempted from federal regulations for the protection of human subjects by the institutional human subjects division because existing data without personal identifiers were used. For the selection of cases and controls, individuals with specific IgE but no history or presence of allergic symptoms ($n = 71/1088$), individuals without detectable specific IgE but history or presence of allergic symptoms ($n = 43/1088$), and individuals with sIgE levels between 50 kU/L and 100 kU/L ($n = 168/1088$) were excluded.

Three criteria were used to define an atopic individual: (1) a positive history of allergy (allergic rhinitis, atopic dermatitis, asthma, food allergy) and/or physical signs of atopy (conjunctivitis, hay fever, rash or asthma); (2) total serum levels (sIgE) ≥ 100 kU/L; and (3) detectable corresponding specific IgE antibodies as determined by fluoro-allergosorbent assay (≥ 0.35 kU/L). Cases were a sample ($n = 60$) of the 283 blood donors who met all three criteria. As spare sera samples with high sIgE levels had been used in another study [23], the cases were not a random sample of all such individuals.

Controls were a random sample ($n = 60$) of all blood donors without history of allergy and physical signs of allergic symptoms, in combination with negative specific IgE screening tests for common food- and aeroallergens ($n = 691$) and with sIgE levels below 50 kU/L. A cut-off level for sIgE of 50 kU/L was chosen because serum levels of atopic and nonatopic individuals overlap to a considerable extent [24]. One control with sIgE levels > 50 kU/L was selected in error and subsequently excluded from the analysis.

Laboratory testing

Determination of IgE, eosinophil and leucocytes

Total sIgE levels from serum specimens were tested in duplicates as previously described [22] at the Institut für Immunologie und Transfusionsmedizin in Lübeck, Germany by means of an enzyme immunoassay using the CAP-RAST system (Pharmacia, Freiburg, Germany). For the purpose of this study, sIgE levels above the calibration limit were coded as ≥ 2000 kU/L.

Specific sIgE levels were determined at the Institut für Immunologie und Transfusionsmedizin in Lübeck, Germany. Specific IgE antibodies against common aeroallergens (sx1: weeds, grasses, trees, cat and dog epithelia, *Cladosporium* sp. and *Dermatophagoides* sp.) and food allergens (fx5: egg white, milk, flour, fish, peanut, soybean) were tested by the multiscreen CAP-RAST system (Pharmacia, Freiburg, Germany) [22]. Samples which were negative in the multiscreen allergen tests despite allergic symptoms were screened for antibodies against insect venom, penicillin and special food- or aeroallergens (Pharmacia, Freiburg, Germany).

Eosinophil and leucocyte counts from venous blood were determined by an automatic cell counter (Coulter T-660, Coulter Electronics Limited GmbH, Krefeld, Germany) as described earlier [22].

Determination of sCD30

sCD30 levels were determined applying a sandwich enzyme-linked immunosorbent assay (ELISA) at the Institut für Pathologie, Freie Universität Berlin, Germany. The ELISA (DAKO, Glostrup, Denmark) is based on two non-competitive monoclonal antibodies which are directed against two different epitopes of the soluble CD30 molecule. The standard curve was prepared from six CD30 calibrators. The concentration of the sCD30 in the sera was determined by interpolation and averaging of values detected in duplicate samples. The reference reagent used to determine the standard curve was originally prepared from an extract of 10^6 cells of the HUT102 cell line which was arbitrarily assigned the value of 1000 U/mL [4,6]. The reproducibility of the assay as assessed by evaluation of the coefficient of variation (CV) was estimated to be $\leq 8.5\%$ by up to five duplicate determinations for five separate runs of four specimens (Dako). The detection limit of the assay was estimated to be 1 U/mL (± 2 SD) by determination of the mean absorbency of 20 measurements of the 0 U/mL standard [9]. The mean sCD30 value ($\pm$ SEM) from 142 healthy blood donors was 4.6 ± 0.4 U/mL with all individual values being < 20 U/mL [7].

Quality control

Serum from collected blood samples was prepared within 4 h. Maximum time before freezing was 90 min at room temperature or 4 h at 4°C . Serum specimens were aliquoted and stored at -20°C without prior thawing and shipped on dry ice. sCD30 levels were determined by a blinded technician from random samples with the same proportions of cases and controls.

Samples were each tested on two occasions. The average of duplicate measurements from the first occasion was combined with the average of duplicate measurements

from the second occasion and the mean of two tests reported. In seven subjects who were failed to be tested twice, the first measurement was chosen for analysis.

Test-retest reliability of the continuous and the binary sCD30 variable (< 20 U/mL vs ≥ 20 U/mL) was assessed in the 114 subjects with repeated measures. The intraclass correlation coefficient of the repeated sCD30 measurement was used to estimate the reliability coefficient from the one way ANOVA [25]. The sCD30 data were transformed (common base 10 logarithm) to meet the assumptions of normal distribution and equal variance. Normality was checked with a normal probability plot. Reliability of the average measurement was determined by the Spearman-Brown formula [25].

Kappa (κ) with a 95% confidence interval (CI) was calculated as an estimate of the reliability coefficient [24] of the binary sCD30.

Data analyses

Variables were first investigated individually and by stratified analysis and then entered into a multiple logistic regression model. Data were analysed with the software packages SPSS (SPSS Inc., Chicago, IL, USA) and EGRET (Statistics and Epidemiology Research Corp., Seattle, WA, USA). Relative risk estimates (odds ratio [OR]) and 95% CI were calculated from logistic regression coefficients. A two-sided α -level of 0.05 was considered significant. Subjects with missing data were excluded ($n = 1$ without information on smoking status, eosinophil count and leucocyte count).

Based on the results from previous sCD30 determinations in other diseases [6–10], the exposure based on the sCD30 ELISA was treated as a binary variable (< 20 U/mL vs ≥ 20 U/mL). For the dose model, sCD30 was coded as a binary variable (< 20 U/mL vs ≥ 20 U/mL) and as a grouped dose variable with cut-off levels at 50 U/mL and 100 U/mL ($0 = < 50$ U/mL, $1 = \geq 50$ and < 100 U/mL, $2 = \geq 100$ U/mL). A test for trend was conducted using the logistic regression analogue to the Mantel extension test [26] adding the grouped dose variable to the binary variable.

Gender, age (years), and current smoking status (on average at least one cigarette per day: yes/no) were considered as a priori confounders. Further potentially confounding variables examined were leucocyte counts ($\geq 10,000/\text{mL}$ vs $< 10,000/\text{mL}$), elevated cytomegalovirus (CMV) titre in the blood (yes/no), eosinophil count ($\geq 0.36/\text{nL}$ vs $< 0.36/\text{nL}$), and first degree family history of allergy (yes/no). As a rule, a change by 10% or more from the crude estimates was considered as confounding. Multiplicative two-way interactions between sCD30 as a binary variable and eosinophil count ($\geq 0.36/\text{nL}$ vs $< 0.36/\text{nL}$) or first degree family history of allergy (yes/no) were tested for statistical significance as an examination of effect modification.

For the purpose of this study, allergic reaction towards seasonal aeroallergens (weeds, grass, trees) was considered as seasonal rhinitis, and asthma, allergic reaction towards perennial aeroallergen (cat/dog epithelia, house dust, mite), rash and neurodermatitis were defined as other allergic disorders. Misclassification of the type of atopic disease was a problem because 41.7% of cases reported combinations of allergic disorders such as seasonal allergic rhinitis with asthma. After excluding all cases with multiple allergic diseases ($n=25$), the sample size was too small to further evaluate whether sCD30 was specifically associated with an allergic reaction towards seasonal aeroallergens.

The association between log-transformed sCD30 levels and log transformed sIgE levels as a continuous outcome was analysed by multiple linear regression.

Results

Characteristics of the study population

The distribution of age and gender among controls was similar to the distribution from the cohort from which cases and controls were drawn (52.5% of controls vs 51.1% in the cohort of blood donors were male; mean age in controls 30.6 years vs 31.4 years in the cohort). Table 1 lists characteristics of the study population. A greater proportion of cases than controls were male (60% vs 53%) and were in the age category of 25–34 years (45% vs 29%). Distribution of smoking status among cases and controls was similar. Relative to cases, the control group had more individuals with positive cytomegalo virus titre in the blood (25% vs 18%). More cases than controls had a positive family history of allergy (53% vs 12%) and elevated eosinophil counts (71% vs 3%). Atopic disorders in this study were mainly due to seasonal aeroallergen (35%).

Mean sCD30 levels for cases were 75 U/mL (SD 110 U/mL) as compared with 35 U/mL (SD 59 U/mL) for controls. The median sCD30 levels were lower than the mean levels (28 U/mL for cases and 14 U/mL for controls). The ranges of sIgE levels of cases (104– ≥ 2000 kU/L) and controls (2–48 kU/L) reflect the selection criteria for cases (≥ 100 kU/L) and controls (< 50 kU/L). The mean sIgE levels were higher than the median levels (407 kU/L vs 288 kU/L for cases and 15 kU/L vs 12 kU/L for controls). The prevalence of sCD30 levels ≥ 20 U/mL was higher among younger blood donors (Fig. 1).

Test–retest reliability

The reliability of the repeated sCD30 measurements was high (intraclass correlation coefficient = 0.73) corresponding to an estimated reliability of the average of 0.84. The

reliability of the binary sCD30 measure (< 20 U/mL vs ≥ 20 U/mL) was good ($\kappa = 0.63$, 95%CI 0.49–0.77).

Soluble CD30 levels as a predictor of atopic disorders

Elevated sCD30 levels (≥ 20 U/mL) were more prevalent in cases (65.0%) than in controls (32.2%) corresponding with an almost fourfold increased risk associated with sCD30 levels ≥ 20 U/mL as compared with sCD30 levels < 20 U/mL (Table 2). The results obtained from logistic regression were in agreement with the results from cross-tabulation. Smoking, age, sex and CMV titre were not confounders of the relationship between sCD30 and atopic disorders. In addition, none of these covariates was a significant predictor of case status. After controlling for smoking, age and gender, the coefficient of binary sCD30 and its significance decreased only slightly. The age, gender and smoking-adjusted OR associated with sCD30 ≥ 20 U/mL was 3.7 (95% CI 1.7–9.1).

Relationship between sCD30 levels and atopic disorders

Based on categories derived from previous evaluations of sCD30, the linear trend test, after adjusting for age, sex and smoking, was not significant (Table 2; $P = 0.57$).

Total serum IgE levels as a predictor of sCD30 levels

The relation between sIgE and sCD30 levels is shown graphically in Fig. 2. Log sIgE levels were a moderate but significant linear predictor of log sCD30 levels. Adjusted for age, sex and smoking, log sCD30 levels increased with log sIgE levels, 0.19 log U/mL per log kU/L sIgE (95% CI for the slope 0.12–0.26 log U/mL). When cases and controls were evaluated separately, log sIgE levels did not significantly predict log sCD30 levels in either group.

Interaction of sCD30 levels and eosinophil counts

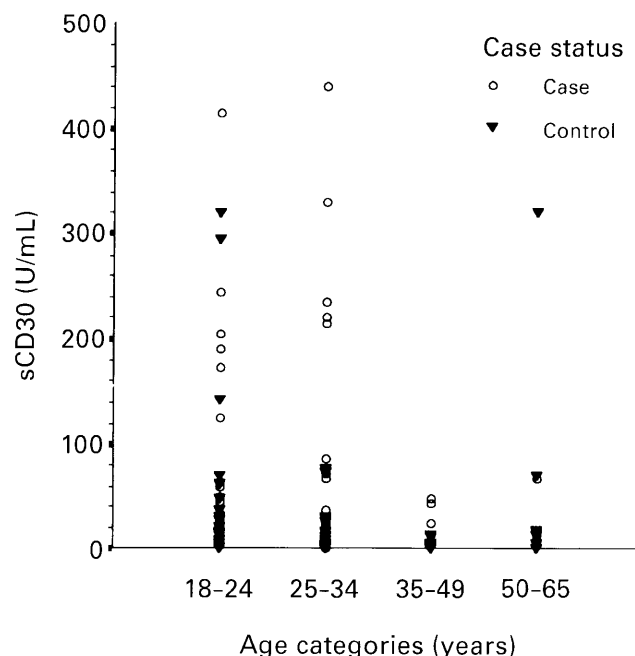
Elevated eosinophil counts were a significant predictor of case status. After adjusting for age, sex and smoking, elevated eosinophil counts were associated with a 11.7-fold increased risk (OR 11.7, 95% CI 2.5–55.1). Eosinophil counts were a weak confounder of the association between sCD30 and case status. When eosinophil counts were additionally controlled, the relative risk of atopic disorder associated with elevated sCD30 levels decreased from an OR of 3.7 to 3.2. The interaction term of sCD30 binary and eosinophil count was not significant.

Interaction of sCD30 levels and family history of allergy

Family history was a very strong and significant predictor of case status. Adjusted for age, sex and smoking, positive

Table 1. Selected subject characteristics

Characteristic	Number (%)	
	Cases <i>n</i> = 60	Controls <i>n</i> = 59
<i>Gender</i>		
Male	36 (60.0)	31 (52.5)
Female	24 (40.0)	28 (47.5)
<i>Age (years)</i>		
18–24	24 (40.0)	24 (40.7)
25–34	27 (45.0)	17 (28.8)
35–49	5 (8.3)	11 (18.6)
50–65	4 (6.7)	7 (11.9)
<i>Smoking</i> ^{1,2}		
No	35 (59.3)	37 (62.7)
Yes	24 (40.7)	22 (37.3)
<i>CMV titre in blood</i>		
No	49 (81.7)	44 (74.6)
Yes	11 (18.3)	15 (25.4)
<i>Family history of allergy</i>		
No	28 (46.7)	52 (88.1)
Yes	32 (53.3)	7 (11.9)
<i>Eosinophil count</i> ¹		
<0.36/nL	17 (28.8)	57 (96.6)
≥0.36/nL	42 (71.2)	2 (3.4)
<i>Leukocyte count</i> ¹		
≥10,000/mL	2 (3.4)	2 (3.4)
<10,000/mL	57 (96.6)	57 (96.6)
<i>Allergy treatment</i> ³		
No	58 (96.7)	59 (100.0)
Yes	2 (3.3)	0 (0.0)
<i>sIgE (kU/L)</i>		
0–50	0 (0.0)	59 (100.0)
100–149	8 (13.3)	–
150–199	11 (18.3)	–
200–299	12 (20.0)	–
300–399	7 (11.7)	–
400–499	8 (13.3)	–
>500	14 (23.3)	–
<i>Specific IgE antibodies</i>		
Not detectable	0 (0.0)	59 (100.0)
Common aeroallergens ⁴	57 (95.0)	–
Common food allergens ⁵	1 (1.7)	–
Other ⁶	2 (3.4)	Not tested
<i>Allergic symptoms</i>		
None	0 (0.0)	59 (100.0)
Seasonal allergic rhinitis	22 (36.7)	–
Perennial allergic rhinitis	7 (11.7)	–
Asthma	3 (5.0)	–
Atopic dermatitis	3 (5.0)	–
Multiple allergens ⁷	25 (41.7)	–

**Fig. 1.** Distribution of sCD30 levels among cases and controls by age.

family history was associated with a 8.6-fold increased risk (OR 8.6, 95% CI 3.3–22.5). If family history was negative, sCD30 levels ≥ 20 U/mL were a strong predictor of case status (OR 4.2, 95% CI 1.5–11.9). If family history was positive, the association between sCD30 levels ≥ 20 U/mL and case status was weaker (OR 2.5, 95% CI 0.5–13.3). The difference could be due to chance.

Discussion

This is the first report on a controlled study evaluating the biological marker sCD30. A significant difference in sCD30 levels between atopic and nonatopic blood donors was demonstrated. The reliability of the average of two sCD30 determinations was high. After categorizing the continuous sCD30 measurements into two categories with a cut-off level of 20 U/mL, the reliability decreased but the agreement was still good [26]. When the authors further examined

¹ One case with missing value excluded. ²Current smoking status (on average at least one cigarette per day: yes/no). ³Desensibilization with allergen. ⁴sx1: seeds, grasses, trees, cat and dog epithelia, *Cladosporium*, *Dermatophagoides*. ⁵fx5: egg white, milk, flour, fish, peanut, soybean. ⁶Special IgE antibodies towards mites (*n* = 1), and mites and weeds (*n* = 1). ⁷Various common aeroallergens (*n* = 21), seasonal allergic rhinitis together with atopic dermatitis (*n* = 2), seasonal allergic rhinitis together with asthma (*n* = 2).

Table 2. Estimated odds ratios (OR) and 95% confidence intervals (CI) for the association between sCD30 and atopy¹ and for the relationship between sCD30 levels and atopy

sCD30 (U/mL)	Cases <i>n</i> (%), <i>N</i> = 60	Controls <i>n</i> (%), <i>N</i> = 59	Crude OR (95% CI)	Adjusted ² OR (95% CI)
<i>Association between sCD30 and atopy</i>				
0–19	21 (35.0)	40 (67.8)	1 (–)	1 (–)
>20	39 (65.0)	19 (32.2)	3.9 (1.8–8.4)	3.7 (1.9–9.1)
<i>Relationship between sCD30 levels and atopy</i>				
0–19	21 (35.0)	40 (67.8)	1 (–)	1 (–)
20–49	20 (33.3)	10 (20.0)	2.8 (1.1–7.2)	2.6 (1.0–6.9)
50–99	7 (11.7)	5 (8.5)	2.0 (0.6–7.3)	1.9 (0.5–7.0)
>100	12 (20.0)	4 (6.8)	5.0 (1.5–17.3)	4.9 (1.4–17.5)

¹sIgE $\geq$ 100 kU/L, presence/history of allergic symptoms, and specific IgE response. ²Adjusted for age, sex and smoking; one case excluded because of missing value for smoking.

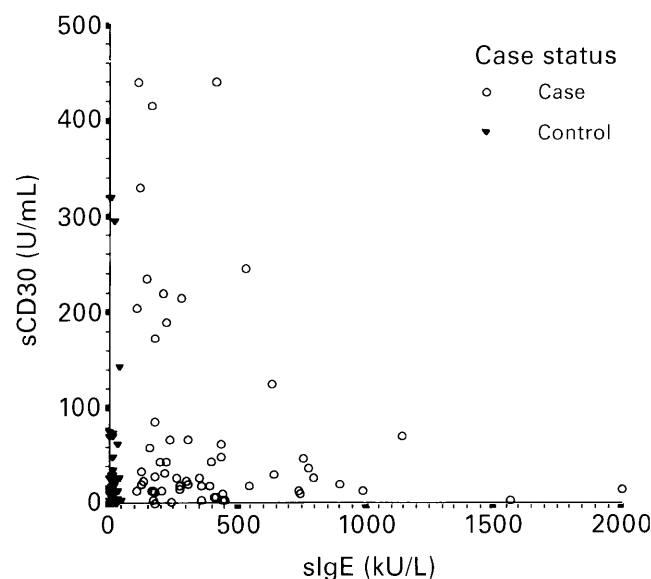
whether increasing levels of sCD30 above 20 U/mL were related to an increasing risk of atopy, no significant trend in risk could be established.

The case group was not a random sample of all eligible atopic blood donors and subjects with severe atopic symptoms were not permitted to donate blood. Selection bias would occur if the relation between sCD30 levels and atopy was different in the selected cases from that in subjects with severe allergic symptoms or in subjects whose sera were no longer available because they had been used in a previous study [23]. There is no reason to assume that the relationship found in the current study would be overestimated: (a) the prevalence of sCD30 among the atopic blood donors selected as cases (65%) was similar to that observed (67%) in the pilot study of 24 patients with uncharacterized allergies from another clinic; (b) selection of serum samples from atopic blood donors in the previous study on sCD23 [23] was not based on either sIgE or sCD30 levels; and (c) the level of sIgE is not a stable marker for subjects with atopic disorders, and in this study no linear association was seen between sCD30 and sIgE. If the missing sera had particularly high sIgE levels and these subjects were individuals with atopic dermatitis who had high sCD30 levels, the current findings may in fact underestimate the association between sCD30 and atopic disorders. The risk estimate would also be biased towards the null if atopic subjects suffering from severe allergic symptoms had particularly high sCD30 levels.

Due to the reimbursement procedure for blood donors in Germany, the sample from this study is not representative. The prevalence of elevated sCD30 levels among controls, particularly in the youngest age group, was much higher than anticipated which should result in an underestimation of the true relative risk. As CD30 is expressed after viral stimulation [5–7,19] and subjects with HIV, hepatitis C,

hepatitis B, and CMV were excluded, a high prevalence of a virus such as EBV [5,16] might be an explanation for the elevated sCD30 levels among the younger age groups of blood donors. In previous studies where sCD30 levels among controls were considerably lower, the population base for the selection of controls was not described [5–14,28–33]. Few controls with very high sCD30 levels were also reported by Bengtsson [29] and co-workers. Information on the distribution of sCD30 levels in a larger population-based sample and a random sample of patients attending an allergy clinic are needed to further evaluate the positive predictive value of the sCD30 ELISA in the target population.

The presence of elevated sCD30 serum in atopic individuals supports initial *in vitro* findings [19]. After the

**Fig. 2.** Scatter plot of sCD30 levels and sIgE levels.

conduct of this study, the presence of CD30 [31,32,34], and elevated sCD30 levels [28–33] were reported in patients with dermatitis [28–31,34], asthma [32], asthma and/or allergic rhinitis [33] but not in patients with uncharacterized respiratory atopic disorders [30]. The presence of cellular [32,34] and soluble [28,31] CD30 was related to clinical scores of disease activity in atopic dermatitis [28,31,34] and disease severity in asthma [32]. Yet, no correlation between sCD30 and disease activity was reported in two studies [29,32]. The value of sCD30 as a marker to monitor disease activity in different atopic disorders should be evaluated in a larger longitudinal study where the association of sCD30 levels with onset of disease, symptom scores, and intervention is further investigated.

About 42% of cases reported combined allergies which is in keeping with 37% as reported in another German study for multiple sensitization to allergens [35]. The association between elevated sCD30 levels and specific type of allergic disease should be evaluated in a larger study on allergic disorders with a diverse spectrum of disease types.

Both environmental and genetic factors are important causes of atopy. Prevalence of family history of allergy among cases in this study was 53% which is comparable with the prevalence (55%) in a study among German school children [36]. Family history did not confound the association of sCD30 and atopy. Elevated sCD30 levels were more prevalent among cases with a negative family history of allergy than cases with positive family history (64% vs 34%) but the results could be due to chance. Stratification on sCD30 in a study on genetic factors of atopy might enhance the ability to detect main effects in subgroups of the study population.

The association of sCD30 with two clinical features of atopic disorders: eosinophilia and elevated sIgE levels was also investigated. Eosinophilia was a strong and significant predictor of atopy. As the risk of atopic disorder associated with sCD30 levels ≥ 20 U/mL was still more than threefold increased after adjusting for eosinophil count, sCD30 seems to be a predictor of atopy which acts mainly independent of elevated eosinophil counts. No linear relationship between sIgE and sCD30 was detected beyond the association between sCD30 and case status. A link between high sIgE levels and CD30 is known from Hodgkin's disease [24]. Consistent evidence of a linear relationship between CD30 and sIgE in atopic disorders is lacking [29,30,33]. A correlation between sIgE and CD30 mRNA was shown in patients with allergic symptoms [33] but no correlation between sIgE and sCD30 was found in patients with atopic dermatitis [29,30].

Results from this study demonstrate that sCD30 can be detected *in vivo* during the course of a conditions where IL-4 is produced. Recent *in vitro* investigations have shown that CD30 is preferentially associated with TH2 responses

because the expression of CD30 by activated T cells is dependent of IL-4 [21]. With this case-control approach and the investigation of disease prevalence, the authors were unable to test for a causal relationship between sCD30 levels and atopic disorders. The results are consistent with the hypothesis that sCD30 is involved in the pathogenesis of atopic disorders. The findings indicate that sCD30 might be a useful marker of atopy in combination with other factors to confirm an atopic aetiology and to monitor disease activity. Moreover, blocking of the CD30/CD30L signalling pathway may lead to new therapeutic approaches.

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