Berger CL, Hanlon D, Kanada D, *et al*: The growth of cutaneous T-cell lymphoma is stimulated by immature dendritic cells. Blood 99:2929–2939, 2002

Braverman IM, Klein S, Grant A: Electron microscopic and immunolabeling studies of the lesional and normal skin of patients with mycosis fungoides treated by total body electron beam irradiation. J Am Acad Dermatol 16: 61–74, 1987

Chang YT, Liu HN, Chen CL, Chow KC: Detection of Epstein–Barr virus and HTLV-I in T-cell lymphomas of skin in Taiwan. Am J Dermatopathol 20: 250–254, 1998

Chu A, Patterson J, Berger C, Vonderheid E, Edelson R: *In situ* study of T-cell subpopulations in cutaneous T-cell lymphoma Diagnostic criteria. Cancer 54:2414–2422, 1984

Dreno B, Celerier P, Fleischmann M, Bureau B, Litoux P: Presence of Epstein–Barr virus in cutaneous lesions of mycosis fungoides and Sezary syndrome. Acta Derm Venereol 74:355–357, 1994

Jumbou O, Mollat C, N'Guyen JM, Billaudel S, Litoux P, Dreno B: Increased anti-Epstein–Barr virus antibodies in epidermotropic cutaneous T-cell lymphoma: A study of 64 patients. Br J Dermatol 136:212–216, 1997

Lee PY, Charley M, Tharp M, Jegasothy BV, Deng JS: Possible role of Epstein–Barr virus infection in cutaneous T-cell lymphomas. J Invest Dermatol 95: 309–312, 1990

Meissner K, Loning T, Rehpenning W: Epidermal Langerhans cells and prognosis of patients with mycosis fungoides and Sezary syndrome. *In Vivo* 7: 277–280, 1993

Association of *TNF −238* and *−308* Promoter Polymorphisms with Psoriasis Vulgaris and Psoriatic Arthritis but not with Pustulosis Palmoplantaris

To the Editor:

Overexpression of tumor necrosis factor (TNF)-α is a central element in the pathogenesis of psoriasis vulgaris (PV) and psoriatic arthritis (PsA); however, the underlying mechanisms are poorly understood. Several recent studies with German patients found an association between the rare *A allele of the G → A single nucleotide polymorphism at position −238 of the *TNFA* promoter and psoriasis, particularly in patients with early disease onset (Arias *et al*, 1997; Hohler *et al*, 1997; Reich *et al*, 1999; Hohler *et al*, 2002; Reich *et al*, 2002). There is also evidence that carriage of the rare *A allele of another G → A single nucleotide polymorphism at position −308 of the promoter is decreased in patients with PsA (Hohler *et al*, 2002), although within this disease subgroup it may be increased in patients with a more progressive course of joint involvement (Balding *et al*, 2003). In light of the possible influence of *TNFA* promoter polymorphisms on cytokine production (Hajeer and Hutchinson, 2001), the increased formation of TNF-α in psoriasis could at least partially be genetically determined.

In this study, *TNFA −238* and *−308* genotypes were analyzed in 239 unrelated patients with PV, 43 patients with pustulosis palmoplantaris (PPP) without concomitant PV, and 135 control subjects according to published methods (Reich *et al*, 2002). All participants were Caucasians and were enrolled at the Department of Dermatology, University of Tartu, Estonia. Patients with PV were considered to have early-onset disease if skin symptoms occurred before 40 y of age, and late-onset disease if age at onset was ⩾40 y (Henseler and Christophers, 1985). Disease severity was assessed at study entry by determination of the psoriasis area and severity index (PASI) (Fredriksson and Pettersson, 1978). PV patients were classified to have concomitant PsA (n = 59) if this diagnosis had been established by an experienced rheumatologist. Clinical deformities of the hands and/or feet consistent with PsA were seen in 23 of these patients, and nine patients had erosions of the hands and/or feet by radiographic assessment. Control subjects were recruited from among medical students, health care personnel, and patients presenting at the dermatological outpatient clinic with mild expression of either facial teleangiectasis or skin tags. The study was approved by the Ethics Review Committee on Human Research of the University of Tartu, and was conducted according to the Declaration of Helsinki protocols. Informed consent was obtained from all participants.

To evaluate deviation from the Hardy–Weinberg equilibrium, observed and expected genotype frequencies were compared by a Monte-Carlo goodness-of-fit test in PV and PPP patients and in controls. Odds ratios (OR) and exact 95% confidence intervals (CI) were calculated to compare genotype frequencies. Carriage rates of variant allele were investigated using the exact test by Fisher. To correct for multiple testing, a hierarchical test strategy was adhered to as described (Reich *et al,* 2002), and the respective nominal p values (p_{nom}) were adjusted according to Bonferroni–Holm (p_{adj}).

Absolute and relative *TNFA* genotype frequencies are shown in Table I. The genotypes were in Hardy–Weinberg equilibrium with the exception of the *TNFA −238* locus in the PPP subgroup (p = 0.0118). *TNFA* genotype frequencies in the control group were similar to those observed in other large European studies (Hohler *et al*, 1997; Reich *et al*, 2002).

Carriage of *TNFA −238*A* was significantly more common among patients with PV than among control subjects (23.8% *vs* 8.9%; OR 3.21 [1.60–6.84], $p_{adj} = 0.0012$), whereas carriage of *TNFA −308*A* was decreased (17.6% *vs* 29.6% in controls, OR 0.51 [0.30–0.86], $p_{adj} = 0.0360$). The latter finding was independent of *TNFA −238* as the difference between patients and controls remained similar after exclusion of *−238*A* positive individuals from the analysis (20.3% *vs* 30.9% in controls).

Abbreviations: CI, confidence interval; OR, odds ratio; p_{adj}, adjusted p value; p_{nom}, nominal p value; PASI, psoriasis area and severity index; PPP, pustulosis palmoplantaris; PsA, psoriatic arthritis; PV, psoriasis vulgaris; TNF, tumor necrosis factor

Table I. Frequencies of *TNFA −238* and *TNFA −308* genotypes in the investigated groups

		Psoriasis vulgaris	PPP	Controls
		(n = 239)	(n = 43)	(n = 135)
	Genotype	n (%)	n (%)	n (%)
TNFA −238	G/G	182 (76.3)	42 (97.7)	123 (91.1)
	G/A	55 (23.0)[a]	0	12 (8.9)
	A/A	2 (0.8)	1 (2.3)	0
TNFA −308	G/G	197 (82.4)	32 (74.4)	95 (70.4)
	G/A	41 (17.2)[b]	11 (25.6)	39 (28.9)
	A/A	1 (0.4)	0	1 (0.7)

[a]Odds ratio = 3.21, 95% confidence interval = 1.60–6.84, nominal p-value = 0.0003, adjusted p-value = 0.0012; *TNF −238*G/A* and **A/A* psoriasis vulgaris *vs* control.
[b]Odds ratio = 0.51, 95% confidence interval = 0.30–0.86, nominal p-value = 0.0090, adjusted p-value = 0.0360; *TNF −308**G/A and ***A/A psoriasis vulgaris *vs* control.
PPP, palmoplantar pustulosis.

Pathogenetic similarities between PV and PPP have recently been shown to include the prominent role of TNF-α (Kitamura *et al*, 1999). But, in contrast to the findings in PV patients, carriage rates of *TNF −238*A* and *TNF −308*A* in patients with PPP were similar to those in the control group (Table I).

In accordance with previous findings, the association between *TNFA −238*A* and PV appeared to be more pronounced in patients with a positive family history (30.3% *vs* 8.9% in controls; OR 4.46 [2.05–10.14], $p_{adj} = 0.0036$) than in those with sporadic disease (19.3% *vs* controls; OR 2.45 [1.13–5.56], $p_{nom} = 0.0155$, $p_{adj} = 0.1860$). Unlike in earlier studies (Reich *et al*, 2002), however, *TNFA −238*A* was likewise increased in patients with early (<40 y) and late disease onset (⩾40 y), and in male and female patients. The decrease of *TNFA −308*A* was seen in patients with early and late disease onset, and seemed to be independent of gender and family history. Due to the smaller number of individuals in the subgroup analysis, not all associations remained significant after correction for multiple testing.

As there is recent evidence that cytokine gene polymorphisms may influence disease severity in PV and PsA (Balding *et al*, 2003; Kingo *et al*, 2003), *TNFA* genotype frequencies were also analyzed separately in patient subgroups with mild (PASI < 12; n = 87) and moderate-to-severe disease (PASI ⩾ 12; n = 152). Genotype frequencies of the *TNFA −238* polymorphism were not significantly different in these groups. But, there was a trend toward a decreased carriage of *TNFA −308*A* in patients with more severe disease. In patients with mild disease, the carriage rate of the **A* allele was 21.8%, and was not significantly different compared with control individuals (29.6%; OR = 0.66 [0.33–1.29], $p_{nom} = 0.2165$), but it was 15.1% in patients with a PASI ⩾ 12 (OR = 0.42 [0.23–0.78], $p_{nom} = 0.0041$, $p_{adj} = 0.1640$ *vs* controls).

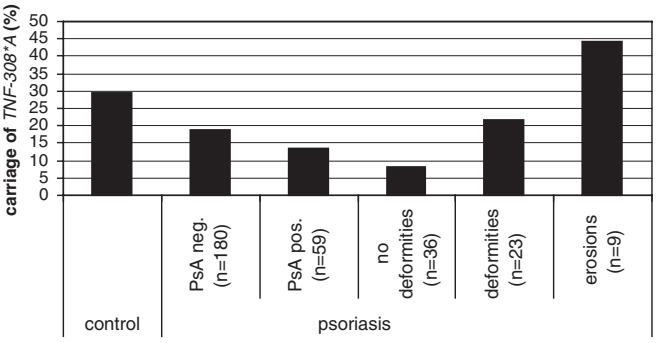

Figure 1
Carriage rate of *TNFA 308*A* is increased in patients with severe psoriatic arthritis. Carriage of *TNFA −308*A* in patients with psoriasis with (PsA pos.) or without (PsA neg.) a confirmed diagnosis of psoriatic arthritis, as well as in patients with PsA with or without clinical deformities or bone erosions of the hands and/or feet. Bars represent the percentage of carriers of *TNFA −308*A* in each of these groups and in control subjects. n, number of individuals investigated in each group.

Increased carriage of *TNFA −238*A* in PV patients compared with controls was more pronounced among patients without arthritis (25.6% *vs* 8.9% in controls; OR 3.52 [1.73–7.62], $p_{adj} < 0.0001$) than among patients with arthritis (18.6% *vs* 8.9%; OR 2.35 [0.87–6.24], $p_{adj} = 0.4376$), while the finding of a decreased carriage of *TNFA −308*A* was more pronounced among patients with arthritis (13.6% *vs* 29.6%; OR 0.37 [0.14–0.89], $p_{nom} = 0.0187$, $p_{adj} = 0.5236$) than among patients without joint involvement (18.9% *vs* 29.6%; OR 0.55 [0.32–0.97], $p_{nom} = 0.0316$, $p_{adj} = 0.7584$). Within the subgroup of patients with arthritis, however, *TNFA −308*A* appeared to be increased in individuals with more severe joint disease, i.e., patients with deformities of the hands and/or feet or radiologically confirmed erosions (Fig 1).

In conclusion, we confirm and extend previous observations on the role of *TNFA* promoter polymorphisms in psoriasis and postulate that (i) carriage of *TNFA −238*A* is associated with PV in Caucasian populations, whereas carriage of *TNFA −308*A* may have a protective effect and is associated with less severe skin disease, that (ii) PPP, at least the PPP variant without concomitant PV, is genetically different from PV, and that (iii) although carriage of *TNFA −308*A* is decreased in PsA compared with controls, within patients with arthritis, it may be a marker of more severe joint involvement. With therapies that antagonize TNF-α becoming a more widely used strategy to treat psoriatic skin and joint symptoms, it will be interesting to see whether the presence of *TNFA* promoter polymorphisms influences the response to treatment. Our study supports the concept of a differential role of *TNFA* polymorphisms in PV and PsA and encourages future investigations in the field.

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References

Arias AI, Giles B, Eiermann TH, Sterry W, Pandey JP: Tumor necrosis factor-alpha gene polymorphism in psoriasis. Exp Clin Immunogenet 14:118–122, 1997

Balding J, Kane D, Livingstone W, Mynett-Johnson L, Bresnihan B, Smith O, FitzGerald O: Cytokine gene polymorphisms: Association with psoriatic arthritis susceptibility and severity. Arthritis Rheum 48:1408–1413, 2003

Fredriksson T, Pettersson U: Severe psoriasis—oral therapy with a new retinoid. Dermatologica 157:238–244, 1978

Henseler T, Christophers E: Psoriasis of early and late onset: Characterization of two types of psoriasis vulgaris. J Am Acad Dermatol 13:450–456, 1985

Hajeer AH, Hutchinson IV: Influence of TNFalpha gene polymorphisms on TNF-alpha production and disaease. Hum Immunol 62:1191–1199, 2001

Hohler T, Grossmann S, Stradmann-Bellinghausen B, *et al*: Differential association of polymorphisms in the TNFalpha region with psoriatic arthritis but not psoriasis. Ann Rheum Dis 61:213–218, 2002

Hohler T, Kruger A, Schneider PM, *et al*: A TNF-alpha promoter polymorphism is associated with juvenile onset psoriasis and psoriatic arthritis. J Invest Dermatol 109:562–565, 1997

Kingo K, Koks S, Silm H, Vasar E: IL-10 promoter polymorphisms influence disease severity and course in psoriasis. Genes Immun 4:455–457, 2003

Kitamura T, Tamada Y, Kato M, Yokochi T, Ikeya T: Soluble E-selectin as a marker of disease activity in pustulosis palmaris et plantaris. Acta Derm Venereol 79:462–464, 1999

Reich K, Mössner R, König IR, Westphal G, Ziegler A, Neumann C: Promoter polymorphisms of the genes encoding tumor necrosis factor-alpha and interleukin-1beta are associated with different subtypes of psoriasis characterized by early and late disease onset. J Invest Dermatol 118: 155–163, 2002

Reich K, Westphal G, Schulz T, *et al*: Combined analysis of polymorphisms of the tumor necrosis factor-alpha and interleukin-10 promoter regions and polymorphic xenobiotic metabolizing enzymes in psoriasis. J Invest Dermatol 113:214–220, 1999

Upregulation of CD44 and Hyaluronate Synthases by Topical Retinoids in Mouse Skin

To the Editor:

CD44 is a ubiquitously expressed cell surface proteoglycan that exists as numerous alternatively spliced and/or post-translationally modified variants (Stamenkovic *et al*, 1991; Screaton *et al*, 1992; Lesley *et al*, 1993; Bennett *et al*, 1995; Zhou *et al*, 1999). CD44 appears to be a major cell surface receptor for hyaluronate [(HA); Aruffo *et al*, 1990; Miyake *et al*, 1990]. In a recent study, we have shown that two major functions of CD44 in the mouse skin are the regulation of keratinocyte proliferation in response to extracellular stimuli and the maintenance of local HA homeostasis (Kaya *et al*, 1997).

HA is a high-molecular-weight linear polymer composed of repeating units of D-glucuronic acid and *N*-acetyl-D-glucosamine and is the principal component of the extracellular matrix (Fraser and Laurent, 1989; Laurent and Fraser, 1992). HA is synthesized by hyaluronate synthase (HAS) at the inner surface of the plasma membrane as long chains by adding sugar residues at the reducing end, and then extruded to the extracellular space (Prehm, 1984). Recently, three distinct yet highly conserved genes encoding mammalian HAS, HAS1, HAS2, and HAS3, have been cloned (Itano and Kimata, 1996; Spicer *et al*, 1996, 1997). *In vitro* studies demonstrated that HAS enzymes are distinct from each other in enzyme stability, elongation rate of HA, K_m values for D-glucuronic acid and *N*-acetyl-D-glucosamine, and average chain length of synthesized HA (Itano *et al*, 1999). All HAS isoenzymes are expressed in the monolayers of rat keratinocytes (Pienimaki *et al*, 2001), whereas human keratinocytes are reported to express only HAS1 and HAS3 (Sayo *et al*, 2002) and mouse skin at least HAS1 and HAS2, HAS1 being expressed more strongly in the epidermis and HAS2 in the dermis (Sugiyama *et al*, 1998).

It has been shown that HA was selectively stimulated by retinoids in porcine epidermis (King, 1984), hairless mouse skin (Margelin *et al*, 1996), and human epidermis in skin organ culture (Tammi *et al*, 1989). The mechanism of this stimulation is not known. CD44 and HA expression show a close correlation in human epidermis (Wang *et al*, 1992) and rat epidermal keratinocyte organotypic cultures (Pasonen-Seppanen *et al*, 2003). The effect of retinoids on CD44 and HAS expression, which might contribute to retinoid-induced HA stimulation, has not been studied.

In this study, we analyzed the effect of a topical retinoid, retinaldehyde (RAL), on the protein and RNA expression of CD44 and the RNA expression of HAS1, HAS2, and HAS3 in mouse skin. Pilot observations were also made in human skin on the CD44 protein expression. The experiments described in the article were approved by the Ethical Commission on Animal Experimentation of the University of Geneva and the Cantonal Veterinary Office of Geneva. The Guidelines on Animal Experimentation of the Cantonal Veterinary Office of Geneva in accordance with the Federal Legislation of Swiss Confederation were followed.

Immunostaining of vehicle-treated back skin of SKH1 hairless mice revealed the standard expression of CD44 in

Abbreviations: HA, hyaluronate; HAS, hyaluronate synthase; RAL, retinaldehyde