Lymphocyte imbalance in vitiligo patients indicated by elevated CD4+ /CD8+ T-cell ratio

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Received November 12, 2008, accepted after revision February 6, 2009

Lymphozytendysbalance bei Vitiligo Patienten – Hinweis durch erhöhtes Verhältnis von CD4- zu CD8 T-Zellen


Bei den meisten Patienten mit Vitiligo erwies sich die Balance von zytotoxischen Suppressorzellen und Helfer T-Zellen im peripheren Blut als gestört, was zu einer Verteilungsstörung der T-Zell-Subtypen am intrakutanen Locus des autoimmunen Melanozytenverlusts führen könnte.

Schlüsselwörter: Vitiligo, Autoimmunologie, Lymphozytenverteilungsstörung, proinflammatorische Cytokine

Summary. Vitiligo is a disorder of pigmentation associated with an autoimmune-mediated loss of melanocytes from the epidermis. Humoral immunity and the involvement of cellular immunity have been investigated in the pathogenesis of vitiligo. We evaluated the role of pro-inflammatory cytokines and lymphocyte fractions in peripheral blood in a cohort of Austrian patients with vitiligo.

Morning blood samples from 40 patients with vitiligo were collected. Twenty-one patients had active and 19 had stable vitiligo disease. All patients were suffering from non-segmental vitiligo at different stages of the disease. Sixteen persons presented with an additional autoimmune thyroid disease. To evaluate a possible involvement of proinflammatory cytokines in vitiligo we measured TNF-RI (soluble tumour necrosis factor receptor I), IL-6 and additionally CIC (circulating immune complexes). We compared these findings to the data from matched normal persons. To investigate the mechanisms of cellular immunity, peripheral blood cell count and lymphocyte subtype analysis by flow cytometry were done.

The patient group median sTNF-RI level was 1.5 ng/ml and median CIC level was 35.2 μg/ml, and no statistically significant differences to the control group were observed. Median IL-6 level in vitiligo patients was 2.7 pg/ml and in the normal range - but higher than the median level of 0.5 pg/ml observed in normal persons (p<0.001). Absolute and relative counts of lymphocyte subtypes were normal. The ratio of CD4+ /CD8+ T-cells had an elevated median value of 2.6 [quartiles 2.0; 3.1]. 61% of the vitiligo patients had a ratio higher than 2.4, which was the normal cut-off point.

In most vitiligo patients the balance of cytotoxic/suppressor and helper/inducer T-cells in peripheral blood is disturbed which might lead to a predominance of T-cell subtypes in the intracutaneous site of autoimmune melanocyte loss.
Key words: Vitiligo, autoimmunity, lymphocyte imbalance, proinflammatory cytokines

Introduction

Vitiligo is an acquired disorder in which white patches of skin result from an autoimmune loss of melanocytes from involved areas. The autoimmune pathogenesis of vitiligo has become a rapidly evolving field of research. A humoral immune reaction has been implicated through the detection of circulating antibodies. Recent research focuses on cytotoxic T-cell immune reaction in the melanocyte destruction [1]. Histological and immunohistochemical studies in perilesional skin suggest the involvement of cellular immunity in vitiligo. T-cell analyses in peripheral blood further support this hypothesis [2]. Data presenting lymphocyte subtype analysis are sometimes contradictory and different pathological conditions are described. This observation is likely to be related to different patient selection criteria based on the study setting.

Different main hypotheses for the pathogenesis of vitiligo exist – strong evidence in favour of the autoimmune hypothesis has been obtained [3]. One major reason is the coexistence of other autoimmune disorders, as autoimmune thyroid disease, pernicious anemia and systemic lupus erythematous [4]. Our patient group includes a high portion of persons with associated autoimmune disease and therefore most likely patients whose vitiligo development is related to immune mediated processes. The aim of the study is to detect etiologically relevant mechanisms of cellular immunity in peripheral blood common to the patient group.

Methods

Patients
We collected morning blood samples from 32 women and 8 men throughout 2002 at the General Hospital Linz. Mean age was 52 ± 18 years, 21 patients had active and 19 had stable vitiligo disease. All patients were suffering from non-segmental vitiligo at different stages of the disease. Sixteen persons presented with an additional autoimmune thyroid disease (11 with Hashimoto’s thyroiditis and five with Graves’ disease, all with levels of fT4 and fT3 in the normal range with or without medication). Three had pernicious anemia and there was one patient with insulin dependent autoimmune diabetes mellitus and one patient with autoimmune hepatitis. Organ-specific autoimmune antibodies were measured in these patients. To evaluate the possible involvement of proinflammatory cytokines in vitiligo, we measured sTNF-RI, IL-6 and additionally CIC. We compared these findings to the data from healthy persons matched for age, sex and number. For further evaluation of involvement of cellular immunity in vitiligo disease, we analyzed blood cell count and lymphocyte subfractions by flow cytometry in peripheral blood. Data of CMV infection parameters [5] and endocrine evaluation including melanotropins [6] in the same patient cohort have been presented elsewhere.

The study has been approved by the local ethic committee.

Laboratory methods
Serum levels of sTNF-RI and IL-6 were analyzed by automated enzyme immunoassays (EIA), the TNF-RI EASIA-CB and the IL-6 using EASIA-CB (BioSource, Fleurus, Belgium) on the Cobas Core II analyser (Roche, Vienna, Austria). According to the manufacturer’s guidelines, the detection limits of sTNF-RI and IL-6 were 0.1 ng/ml and 1.5 pg/ml, respectively. Both EIAs exhibited a 100% specificity for human sTNF-RI and human IL-6.

The assays used for the assessment of the standard thyroid parameters are presented in Tab. 1. All assays were routinely performed according to the manufactures’ instructions.

Statistics
Values are presented as mean ± standard deviation or as median with quartiles in brackets. Adjustment to normal...