Value of Interleukins 4, 10, 12, 18 and Interferon Gamma in Acute Versus Chronic Atopic Dermatitis

Samar A.M. Salem, M.D.*, Heba M. Diab, M.D.*, Ghada Fathy, M.D.*, Lamia M. Obia, M.D.* and Shereen B. El Sayed, M.D.†

*Department of Dermatology and Venereology and †Department of Medical Microbiology and Immunology, Faculty of Medicine, Ain-Shams University, Egypt.

Background. Atopic dermatitis (AD) is a chronic inflammatory skin disease with 2 phases, acute and chronic. No therapeutic attempts has yet been tried to target these phases rather than treatment according to severity grade. Animal and in vitro studies pointed to interleukin (IL)-18 as key player in the pathogenesis of AD and the switch between its two phases.

Objective. To evaluate serum IL-18 among other T helper (Th) cytokines (IL-4, IL-10, IL-12, and interferon (IFN)-γ) in acute and chronic AD.

Patients and methods. Thirty AD patients classified into acute and chronic groups, in addition to 20 control subjects were included. Serum IL-4, IL-10, IL-12, IL-18, IFN-γ and serum IgE were estimated by ELISA and the results were interpreted.

Results. IL-12, IL-18 and IFN-γ levels were 2 - 4 folds more in chronic compared to acute AD patients, whereas other studied Th2 cytokines, IL4 and IL10, were significantly higher in acute compared to chronic AD patients. IL-18 correlated positively with disease severity.

Conclusion. IL-12 and IL-18 has a pivotal role in acute and chronic AD. IL-18 measurement is a valuable tool for assessment of disease severity.

(J Egypt Women Dermatol Soc 2010; 7: 56 - 60)

Keywords. Atopic dermatitis, interleukins, interferon gamma

Patient and Methods

This study included 30 AD patients and 20 healthy volunteers as controls. Patients were diagnosed clinically based on UK Working Party’s Diagnostic Criteria. All subjects did not receive any topical treatment for 2 weeks or systemic treatment for...
three months prior to the study and did not have manifestations of other dermatological or obvious medical, autoimmune or infectious diseases. An informed written consent was obtained from the subjects or their legal guardians before their contribution in this study. All subjects of the study were subjected to thorough history taking, general and dermatological examination, blood sampling for quantitative estimation of serum IL-4, 10, 12, 18, IFNγ and total serum IgE. The disease severity was assessed using the SCORAD index.14

Patients were classified into two main groups: acute AD and chronic AD according to the morphology of lesions.11

Estimation of serum IL-4, IL-10, IL-12, IL-18 and IFN γ and IgE

Five milliliters of blood were collected from all subjects in sterile tubes, left to clot, centrifuged and serum was transferred to Eppendorf tubes. The tubes were labeled and stored at -20°C until quantitative assay of IL-4, 10, 12, 18 and IFN-γ was done. Quantikine colorimetric sandwich ELISA immunoassay kits (R&D Diagnostic Minneapolis, USA) were used for particular quantitative determination of serum levels for each of these cytokines; IL-4, IL-10, IL-12, IL-18 and IFN-γ and concentrations were expressed in pg/ml. Undetected cytokine levels were set at the minimum detectable concentrations for kits (8 pg/ml). Quantitative determination of serum IgE was also done by ELISA immunoassay kit (Biocheck, California, USA) and IgE concentrations were expressed in IU/ml. Normal values were considered up to 90 IU/ml.

Statistical analysis

Analysis of data was done by IBM computer using statistical program for social science (SPSS) version 12. Quantitative variables were expressed as mean ± standard deviation (SD). Statistical tests included Chi-square test and Spearman correlation coefficient rank test (r) which was used to rank different variables against each other either positively or inversely. Unpaired t-test was used to compare two independent groups as regards quantitative variables in parametric data (SD < 50% mean). Results were considered significant when p value was ≤ 0.05.

RESULTS

This study included 30 atopic patients, 16 females (53.3%) and 14 males (46.7%). Their age ranged from 6 – 20 years with a mean of 7.54 ± 2.59 years. SCORAD index ranged between 20.4 and 83.5 with a mean of 44.49 ± 23.08. Twelve patients (40%) were categorized as acute AD while 18 patients (60%) were diagnosed as chronic AD. Twenty clinically free volunteers served as controls. They were 11 females (55%) and 9 males (45%). Their age ranged from 10 - 20 years with a mean of 9.44 ± 3.7 years. Both patients and controls were sex and age matched with no statistically significant difference in-between (p > 0.05).

A statistically significant positive correlation was found between the age of patients and disease severity by SCORAD index (r = 0.69, p < 0.0001). As regards the family history of allergic conditions, the overall percentage of AD patients with positive maternal family history was 80% while 60% had positive paternal family history and 33.3% had both maternal and paternal family history of atopy and/or asthma. Although the percentage of patients with combined paternal and maternal positive family history of atopy and/or asthma was more in the chronic AD group (38%) compared to that in the acute AD group (25%) (7/18, 3/12 respectively), the difference was not statistically significant (p > 0.05).

All studied ILs, IFNγ and IgE were statistically significantly higher in patients compared to controls (Table 1). On comparing cytokine patterns in acute and chronic groups of AD, there was a statistically significant increase in the mean values of both IL-4 and IL-10 in acute AD patients (p < 0.05). These values were three to four times less in the chronic AD group. As regards Th1 cytokines, there was three times higher mean value of IL-12; two to three times higher mean value of IL-18 and four times higher mean value of serum IFN-γ with a statistically significant difference in chronic compared to the acute AD groups (Table 2).

A high statistically significant positive correlation was found between serum IL-18 and SCORAD index among studied cases (r = 0.71, p < 0.001) (Figure 1), while no significant correlation was detected between serum IgE and SCORAD (r = 0.17, p > 0.05). Comparison between levels of serum IL-18 in patients with normal serum IgE versus those with high serum IgE revealed no statistically significant difference (Table 3).

Figure 1. Scatter diagram showing the positive correlation between SCORAD index and serum IL-18.
DISCUSSION

Atopic dermatitis is a multifactorial genetically based disease that affects 10 to 15% of children in many parts of the world and constitutes a significant burden to patients and their families. The pathogenesis of AD is considered a combination of both IgE mediated and delayed type hypersensitivities. There is compelling evidence that during the course of AD, the cytokine profile changes from Th2 pattern in acute phase to Th1 cytokine pattern later on.

IL-18 has diverse biological actions depending on its immunological environment. Animal studies found that IL-18 injection induced IFN-γ production and down regulated both IL-4 and IgE in the presence of IL-12. Thus, it was concluded that IL-12 and IL-18 have a role in the switch from Th2 to Th1 cytokine pattern.

IL-18 has diverse biological actions depending on its immunological environment. Animal studies found that IL-18 injection induced IFN-γ production and down regulated both IL-4 and IgE in the presence of IL-12. Thus, it was concluded that IL-12 and IL-18 have a role in the switch from Th2 to Th1 cytokine pattern.

In this study, serum IL-18 levels in acute and chronic AD patients were evaluated among other main T helper cell cytokines. To the best of our knowledge, this comparison was not previously investigated in humans in vivo. Moreover, new trends of immunologic targeted therapy are now under trial and could be the solution to AD patients’ distress. The current study also aimed at evaluating the usefulness of serum IL-18 as an objective marker to assess severity of AD rather than the roughly used serum IgE. Serum IgE lacks specificity as it might be elevated in many instances not related to skin diseases as parasitic infestations. At the same time, normal serum IgE level may be seen in some AD patients.

In the current study, the severity of AD was higher in older patients. This may be related to more exposure to food and aeroallergens with advancing age. Another observation in our study was that 80% of AD patients had positive maternal family history and 60% had positive paternal family history further supporting the genetic background of AD. In patients with both maternal and paternal background, there was an earlier age of disease onset and higher SCORAD index.

These results are in agreement with Betrani and Boguniewicz who reported that the strongest risk factor for AD is the parental history of atopy.

As regards serum IL-18, the present study demonstrated that the mean value of serum levels of IL-18 was significantly higher in atopic patients in comparison to healthy controls which was also the case with other assessed cytokines including IL-4, 10, 12 and IFN-γ. This supports the role of both Th1 and Th2 cytokines in AD which is in...
agreement with other studies implicating the later cytokines in the pathogenesis of AD.\(^5\,2,3,9\)

On comparing cytokine patterns in acute versus chronic phases of AD, there was a statistically significant higher mean values of both serum IL-4 and IL-18 in acute compared to chronic AD patients while, on the other hand, serum IL-12, 18 and IFN-\(\gamma\) were statistically significantly higher in the chronic AD group. As Th2 cytokines predominate in acute AD and Th1 cytokines predominate in the chronic phase, the present results provided a laboratory support for the clinical classification of patients into acute and chronic AD.

Considering the fact that both IL-12 and IL-18 are inducers for IFN-\(\gamma\) production, as concluded from mice studies, with a more potent effect of IL-18 on such induction\(^4\,5\,20\), the above mentioned results support this effect in human with a possible subsequent induction of chronic phase of AD. In the current study, despite the expression of IL-12, IL-18 and IFN-\(\gamma\) in the acute AD group (without clinically manifesting chronic lesions) and taking into account their statistically significant higher values in the chronic AD group, it seems that IL-12 and IL-18 must reach a certain high serum threshold and/or needs a specific associated cytokine milieu to conduct the switch from acute Th2 predominant phase to chronic Th1 predominant phase.

From the previous findings and explanations, a possible suggested sequence of events in AD would be the initiation of acute phase of the disease in genetically predisposed patients with allergen exposure by Th2 cytokines with increased serum IgE. At that stage, serum IL-18 is significantly elevated. However, on reaching certain serum threshold (23 fold increase), and in presence of IL-12, it induces the switch from Th2 to Th1 cytokine pattern with subsequent production of IFN-\(\gamma\) and induction of the chronic phase.

Considering the above mentioned data, it seems appealing to try using anti-Th2 cytokines in the acute phase AD and IL-18 antagonists in chronic AD. Indeed, some animal studies found that IL-18 treatment substantially inhibited the elevation of serum IgE, serum IL-4 levels and dermatitis through increasing IFN-\(\gamma\)\(^7\,21\). Such increase in IFN-\(\gamma\) becomes rapidly reversible once its stimulating cytokine signal is abolished\(^8\). This supports the suggestion of its use to abort acute AD without triggering the chronic phase.

This work showed a statistically significant positive correlation between IL-18 and SCORAD index which emphasizes the clinical relation of serum IL-18 with the severity of inflammatory response in AD. The present results support the previously published data by Hon et al.\(^5\) as regards the significant positive correlation between serum IL-18 and SCORAD index\(^5\,14\,22\).

Results of the current study supported the previously published data\(^9\,11\) concerning the significantly higher values of serum IgE in atopic patients than in healthy controls. However, in this study, there was no statistically significant correlation between SCORAD index and serum IgE values. This was supported by other studies indicating that serum IgE has a poor value in assessment of AD severity\(^23\,24\). The present study also found no statistically significant difference between the mean values of serum IL-18 in AD patients with normal serum IgE and those with high levels of serum IgE. This denotes that serum IL-18 is a more valuable objective factor in assessing AD severity and can be used as a marker to follow up atopic patients. The above results also imply the involvement of serum IL-18 in pathogenesis of both intrinsic AD (with normal serum IgE) and extrinsic AD (with increased serum IgE)\(^12\,23\,24\).

**Conclusion**

The present results confirmed a significant correlation between serum levels of IL-18 and the severity of atopic dermatitis. Measuring serum IL-18 concentration is suggested as an objective practical method for assessment of AD severity as it proved to be more valuable than serum IgE. This work supports the suggested role for IL-18 in the switch from Th2 response in acute AD into Th1 response in chronic AD. The complex interactions between IL-18 and other cytokines in AD need further investigations.

Further studies on larger scales are needed to evaluate the benefits of cytokine targeted therapy, whether IL-18 or its antagonist, in treatment of acute and chronic phases of AD respectively.

**REFERENCES**


