Abstract. Recent data suggest that severe psoriasis is an independent risk factor for chronic renal disease. In the present study, we investigated the role of related-purine derivatives as predictors of renal dysfunctions in patients with psoriasis. A prospective study was conducted on a group of 45 patients with psoriasis vulgaris and 45 control cases, monitored over a 5-year period. Alterations of renal function, albumin/creatinine ratio (ACR, mg/g) and UA/creatinine ratio (UACR, mg/mg) were determined in spontaneous urine samples. The status of related-purine derivatives was evaluated by quantification of uric acid (UA, mg/dl), adenosine deaminase (ADA, UI/mg protein), xanthine oxidase (XO, UI/mg protein) and 8-hydroxy-deoxy-guanosine levels (8-OHdG, ng/ml) in serum samples. Compared to the controls, in patients with psoriasis there was an increase in related-purine derivatives levels, which was demonstrated by the elevated serum levels of UA (5.1±0.4 vs. 5.4±1.0, P=0.066), ADA (0.14±0.08 vs. 0.29±0.12, P=0.052), XO (0.22±0.11 vs. 0.42±0.21, P=0.011) and 8-OHdG (3.1±0.05 vs. 8.3±4.7, P=0.002). The serum levels of related-purine derivatives were associated with the severity of psoriasis. In addition, there was a link between the serum levels of related-purine derivatives and markers of renal impairment. There were positive correlations between 8-OHdG and ACR (r=0.452, P=0.028) and between ADA, XO, UA, 8-OHdG and UACR (r=0.297 and P=0.032, r=0.301 and P=0.002, r=0.431 and P=0.027, r=0.508 and P=0.002) and negative correlations between UA, 8-OHdG and the estimated glomerular filtration rate (r=-0.301 and P=0.036, r=-0.384 and P=0.002). Thus, severe psoriasis is a risk factor for the development of renal disease.

Introduction

Psoriasis is a chronic immunologically mediated dermatosis, affecting 2-3% of the general population (1-3). Multiple mechanisms are involved in its pathogenesis, such as the hyper-reactivity of T-lymphocytes and dendritic cells, excessive inflammatory cytokine synthesis, accelerated epidermal turnover, epidermal hyperproliferation, reduced keratinocyte differentiation, overexpression of angiogenesis and oxidative stress (4-14). The extent of skin involvement is variable, ranging from several psoriatic plaques to generalized forms. The disease evolves with periods of exacerbation and remission (15,16).

It seems that severe psoriasis is an independent risk factor for chronic renal disease (17,18). The mechanisms that mediate kidney failure in patients with psoriasis are controversial. In a retrospective study, it was estimated that patients with psoriasis develop chronic renal disease at a higher percentage compared to controls (5 vs. 2%). Moreover, the risk of kidney disease was higher in the young patients (18). The treatment should be adapted to meet the individual needs of the patients with psoriasis. Non-pharmacological interventions (diet, cessation of smoking and alcohol intake, weight loss, physical exercise) may improve the response to therapy. Potentially nephrotoxic drugs should be used with caution and renal function should be periodically monitored in patients with psoriasis in order to minimize the risk of adverse renal events (13,15,19,20).

In medical literature, there are few studies on the relationship between uremic toxins and the decline of renal function in patients with psoriasis vulgaris. Recent data suggest the role of several serum and urinary markers in the early detection and monitoring of renal disease. The urinary levels of creatinine, albumin, uric acid (UA) and the estimated glomerular
filtration rate (eGFR) can be evaluated, as well as cystatin C, neutrophil gelatinase-associated lipocalin, kidney injury molecule 1, cytokines and chemokines (21). The progressive increase in the level of uremic toxins exerts a negative impact on kidney function (22-25).

Purine derivatives pertain to the class of low molecular weight uremic toxins, which frequently accumulate in the body. Low molecular weight uremic toxins are water-soluble compounds with a molecular weight below 0.5 kDa, and consequently are easily removed by dialysis, and do not exert harmful effects on the body (26,27). The most important uremic toxins classified as purine derivatives are: Products resulted from the degradation of purines (adenosine, inosine, xanthine, guanosine, hypoxanthine, guanine, UA), products of guanosine triphosphate catabolism (neopterin) and oxidative DNA base damage product (8-hydroxy-deoxy-guanosine) (28-30).

The metabolic pathways of purine catabolism imply several phases (31).

The first step involves the transformation of purine mononucleotides (AMP-adenosine monophosphate, IMP-inosine monophosphate, XMP-xanthine monophosphate, and GMP-guanine monophosphate) into purine nucleosides (adenosine, inosine, xanthosine and guanosine), a reaction catalyzed by 5-nucleotidase (E.C.3.1.3.5). Subsequently, conversion of purine nucleosides occurs which frees purine bases (adenine, hypoxanthine, xanthine and guanine), a reaction catalyzed by purine nucleoside phosphorylase (E.C.2.2.2.1). The next step involves the conversion of adenine to hypoxanthine (a reaction catalyzed by adenine deaminase - E.C.3.5.4.4) and then to xanthine (a reaction catalyzed by xanthine oxidase - E.C.1.17.3.2). Guanine is converted to xanthine by guanine deaminase. The final product of purine metabolism is UA, obtained through the enzymatic oxidation of xanthine (a reaction catalyzed by xanthine oxidase) (Fig. 1).

In psoriasis patients, the severity of the skin disease, assessed by the Psoriasis Area Severity Index (PASI), has been analyzed in relation to the prevalence of renal dysfunctions (3,16,18,32,33), purine catabolism (4) and the oxidative stress level (5,6,34). Alteration of purine degradation may be associated with the extension of psoriasis lesions, stimulation of epidermal proliferation and increased DNA peroxidation (6,35-38).

These reactions produce a wide range of reactive oxidants: hydrogen peroxide (H₂O₂), superoxide anion (O₂⁻), hydroxyl radical (OH·), nitric oxide (NO·), peroxynitrite (ONOO⁻), and carbonate radical (CO₃²⁻). Reactive oxygen species and reactive nitrogen species, formed under the action of xanthine oxidase, act on proteins, lipids and nucleic acids, causing damage and cell toxicity (39-42). 8-Hydroxy-deoxyguanosine, a representative metabolite for purine derivatives toxins (43), is a useful indicator for early diagnosis and management of patients with psoriasis (6).

Based on these considerations, we aim to establish characteristic patterns of related-purine derivatives that could represent specific serological markers for the identification of those patients with psoriasis vulgaris at a higher risk of developing renal dysfunctions. In this context, the serum profile of related-purine derivatives was analyzed in psoriasis patients in correlation with the severity of the skin disease and markers of renal impairment. The biomarkers carried out in this study, grouped under the name of related-purine derivatives, include UA (the final product of purine catabolism), adenosine deaminase (ADA, an enzyme which converts irreversibly adenosine and deoxyadenosine to inosine and deoxy-inosine), xanthine oxidase (XO, which catalyzes the oxidation of hypoxanthine to xanthine as well as the oxidation of xanthine to UA) and 8-hydroxy-deoxy-guanosine (8-OHdG, a biomarker of oxidative DNA damage). Renal function was assessed through the serum creatinine level, urinary determinations of eGFR (ml/min/1.73 mp), albumin/creatinine ratio (ACR, mg/g) and UA-creatinine ratio (UACR, mg/mg).

Materials and methods

Study participants. All the study participants provided consent to the use of their biological samples in research studies. The Ethics Committee of “Victor Babes” Clinical Hospital for Infectious Diseases (Bucharest, Romania) approved the study protocol.

A prospective study was conducted on 45 patients with psoriasis vulgaris and 45 control cases, monitored over a 5-year period. Inclusion criteria for the study were: Adults, normolipidemic, normoponderal, with a balanced diet. Exclusion criteria for the study were: Cardiovascular disease, metabolic syndrome, diabetes, anemia, urinary tract infections, nephrolithiasis, leukemia, Lesch-Nyhan syndrome, Wilson’s disease, viral hepatitis, sickle-cell disease, chronic kidney disease, xanthinuria, lead toxicity, treatment with nephrotoxic drugs, folic acid deficiency, pregnant women and breastfeeding women.

The severity of psoriasis was assessed using the PASI score, which assesses the severity of three clinical signs (erythema, thickness, scaling) on a scale from 0 to 4 and the percentage of the skin area involved. The PASI score interpretation was as follows: <7, mild chronic plaque-type psoriasis; 7-12, moderate chronic plaque-type psoriasis; >12, severe chronic plaque-type psoriasis (41).

Changes of glomerular permeability were detected by determining eGFR, ACR and UACR. To assess alterations of renal function, ACR and UACR were determined in spontaneous urine samples. The status of related-purine derivatives was evaluated by quantification of the serum levels of UA (mg/dl), adenosine deaminase (U/mg protein), XO (U/mg protein) and 8-OHdG (ng/ml).

Biological samples were a spontaneous urine sample, preferably first morning urine collected in sterile, preservative-free containers. Urine specimens were centrifuged at 3,000 x g for 10 min at room temperature 20°C. The supernatant was used for measurement of the biological parameters. Venous blood samples (7 ml) were collected in a vacutainer without an anticoagulant and centrifuged at 6,000 x g for 10 min at 20°C. The supernatant was used for biochemical determinations.

Laboratory methods. The determination of creatinine was performed using the colorimetric technique. The method is based on the reaction between creatinine and picric acid in alkaline medium. The absorbance measured at a wavelength of 492 nm is directly proportional to the amount of creatinine in the sample.
The determination of albuminuria was performed via turbidimetric immunoassay using polyclonal antibodies against human albumin. The determination of UA was performed by the colorimetric technique, using the reaction catalyzed by uricase. The concentration of quinonimine resulted from the reaction was measured at a wavelength of 546 nm. The determination of xanthine-oxidase was performed using a spectrophotometer (HumaStar 300; HUMAN Gesellschaft für Biochemica und Diagnostica mbH, Germany, Wiesbaden). Absorbance of the obtained coloured complex was read at a wavelength of 570 nm. The determination of adenosine deaminase was performed via spectrophotometry and the evaluation of the resulting product was measured at a wavelength of 550 nm.

The quantitative determination of hs 8-OHdG (highly sensitive oxidative DNA adduct 8-OHdG) was produced in serum using an enzyme-linked immunosorbent assay (ELISA). The principle of the method is based on the ability of DNA oxidation products to interact with 3,3,5,5-tetramethylbenzidine. The method uses DNA-specific monoclonal antibodies, which cross-react with the oxidative degradation products of DNA (8-hydroxy-guanine, 8-hydroxy-guanosine). The final product of the reaction is colorimetrable via microplate reader (Tecan Austria GmbH, Grodig, Austria) at a wavelength of 450 nm.

Statistical analysis. A comparison of the obtained results between the groups for quantitative variables was performed using the t-test. The correlations between variables were determined by linear regression. The relationship between pairs of two parameters was assessed by Pearson's correlation coefficient (r). We chose a significance level (p) of 0.05 (5%) and a confidence interval of 95% for hypothesis testing.

Results

Patient characteristics. We performed a prospective observational study on 45 patients with psoriasis vulgaris (duration of psoriasis 6.4±3.1 years, PASI 10.4±5.3) and 45 healthy volunteers, who met the inclusion criteria (Table I).

Table I. Characteristics of study participants.

<table>
<thead>
<tr>
<th>Items</th>
<th>Psoriasis vulgaris</th>
<th>Control</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>40.3±11.3</td>
<td>39.4±8.3</td>
<td>0.763</td>
</tr>
<tr>
<td>Male/female ratio</td>
<td>24/21</td>
<td>25/20</td>
<td>0.923</td>
</tr>
<tr>
<td>Smokers/non-smokers ratio</td>
<td>11/34</td>
<td>9/36</td>
<td>0.755</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>20.4±1.3</td>
<td>19.8±1.1</td>
<td>0.899</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>117.3±0.7</td>
<td>115.5±0.5</td>
<td>0.677</td>
</tr>
<tr>
<td>Diastolic blood pressure (mmHg)</td>
<td>72.3±0.2</td>
<td>73.5±0.4</td>
<td>0.546</td>
</tr>
</tbody>
</table>

BMI, body mass index.

Serum determinations. The serum concentration of UA was 5.4±1.8 mg/dl in patients with psoriasis vulgaris and 5.1±0.4 mg/dl in the control group, with statistically insignificant variations between the two groups. There was an important difference between the activity of ADA in patients with psoriasis vulgaris and controls (0.29±0.12 UI/mg protein vs. 0.14±0.08 UI/mg protein, P=0.052). The activity of XO was also significantly increased in psoriasis patients versus controls (0.42±0.21 UI/mg protein vs. 0.22±0.11 UI/mg protein, P=0.011). The serum level of 8-OHdG in patients with psoriasis vulgaris was significantly higher compared to controls (8.3±4.7 ng/ml vs. 3.1±0.05 ng/ml, P=0.002) (Table II).

Urinary determinations. Since the creatinine excretion is a relatively constant variable, the determination of urinary creatinine may be valuable in estimating the renal function. Therefore, the urinary creatinine concentration can be used as a reference parameter for albuminuria and UA. Statistically significant variations were observed for ACR and UACR between patients with psoriasis vulgaris and controls (Table III).
Correlation of serum levels of related-purine derivatives with the severity of psoriasis. Compared to the controls, UA, ADA, XO and 8-OHdG levels were significantly higher in patients with psoriasis with PASI >12 (P<0.05) (Table IV).

Positive correlations between ADA, XO, UA and PASI >12 (r=0.498, P=0.004; r=0.601, P<0.001 and r=0.421, P=0.017) were obtained. There was a strong positive correlation between the serum levels of 8-OHdG and PASI (r=0.406, P=0.008 for PASI ranging from 7 to 12, r=0.782, P=0.000 for PASI >12) (Table V).

Correlations between serum related-purine derivatives and markers of renal impairment. Serum markers of related-purine derivatives in patients with psoriasis were associated with markers of renal impairment. Positive correlations between 8-OHdG and ACR (r=0.452, P=0.028), between ADA, XO, UA, 8-OHdG (r=0.297 and P=0.032; r=0.031 and P=0.002; r=0.431 and P=0.027; r=0.508 and P<0.001) and UACR. Negative correlations between UA, 8-OHdG and eGFR (r=-0.301 and P=0.036; r=-0.384 and P=0.002) were registered (Table VI).

Discussion

In the human body, the progressive increase in the level of uremic toxins exerts a negative impact on all organs, tissues and systems, causing acute and chronic renal dysfunctions, cardiovascular, respiratory and hepatic diseases, atherosclerosis, fibrosis or metabolic alterations (22-25). The presence of UA derivatives is associated with the induction of specific dysfunctions and the normalization of UA levels results in the resolution of the clinical manifestations (26,27,44-46). In terms of chemical structure, uremic toxins are purine, pyrimidine, methylamine, phenyl or indole derivatives, guanidine, polyols, ribonucleosides, peptides, cytokines, advanced glycation end products, advanced lipoxidation end products and reactive carbonyl compounds (26,27,46).

The role played by purine degradation in the pathogenesis of psoriasis is an exciting research topic. The results of the present study on the status of related-purine derivatives in patients with psoriasis indicate changes of the serum level of UA and 8-OHdG and alteration of the enzymatic activities of adenosine deaminase and xanthine oxidase depending on the clinical severity of psoriasis and the stage of renal chronic disease. These results draw attention to the cumulative toxic effect of purine derivatives on the kidney.

In the present study, we obtained slightly increased levels of UA, without statistical significance, in patients with psoriasis versus controls. A significant variation in serum UA levels correlated with the severity of psoriasis. In patients with severe psoriasis, with a PASI score higher than 12, a significantly increased level of UA was obtained compared for both controls and the group of patients with mild or moderate psoriasis. This was also supported by the positive relationship established between serum UA levels and PASI score and between serum UA levels and the results of the tests assessing the renal function in patients with psoriasis.

Stimulation of epidermal proliferation and increased DNA damage may be associated with hyperuricemia, and the normal UA levels could be explained by the selection criteria of patients [normal body mass index (BMI), lack of inflammation, balanced nutritional status]. In addition, the results of this study show that the risk of kidney disease is more evident in patients with severe psoriasis. Our findings are consistent with several reports that have emphasized harmful effects of UA on kidney, the target organ of hyperuricemia (26-28). Data related to the correlation between serum UA levels and the severity of psoriasis are inconsistent. In some studies, elevated serum values were obtained in patients with psoriasis versus controls (38), whereas other studies reported normal UA levels in psoriasis (35,37). Positive correlations between UA levels and the following parameters have been reported: PASI score, cutaneous extension of psoriatic lesions, and BMI (35).

Elevated levels of serum UA promote endothelial dysfunction and renal lesions by decreasing the availability of nitrogen monoxide and inducing oxidative stress. UA-induced endothelial damage could be caused by the reduction in intracellular ATP due to the inactivation of aconitase-2 and enol CoA-hydratase-1, decreased mitochondrial DNA/nuclear DNA ratio, increased mitochondrial calcium, resulting in the
alteration of membrane potential and generation of reactive oxygen species (47). Consequently, endothelial dysfunction can be associated with systemic manifestations (arteriosclerosis, insulin resistance) and renal damage (hypoxia, inflammation, glomerulosclerosis, tubulointerstitial fibrosis) (47,48).

In this study, we revealed that elevated UA levels observed in patients with severe psoriasis were associated with increased xanthine oxidase activity. The serum levels of xanthine oxidase were negatively correlated with eGFR and UACR in urine. The relationship between xanthine oxidase and renal impairment can be explained by the activation of renin-angiotensin system, preglomerular arteriolopathy, induction of oxidative stress and inflammation, and endothelial dysfunctions (49).

The role of xanthine oxidase in the terminal differentiation of keratinocytes may be explained by localization of the enzyme in the granular layer of the epidermis (50). The stimulation of the inflammatory process in human keratinocytes by irradiation with UV rays was correlated with the overexpression of xanthine oxidase and increased production of superoxide (51).

Adenosine is another endogenous purine nucleoside, possibly involved in the pathogenesis of psoriasis. It is thought that adenosine may exert anti-inflammatory and immunomodulatory effects through specific receptors expressed on endothelial cells, leukocytes, mast cells, macrophages, dendritic cells and consequently may limit the extension of psoriatic lesions. Anti-inflammatory effects of adenosine may be achieved by increasing intracellular cAMP levels, modulation of apoptosis, reduction in cytokine synthesis, leucocyte recruitment and immune function.
The relationship between psoriasis and renal disease can be explained by three main mechanisms: Immune-mediated renal damage, drug-related renal damage, and chronic-renal damage (33).

In summary, psoriasis vulgaris can be regarded as a cascade of events that starts from inflammation, oxidative stress and a series of comorbidities. Our study indicates that renal impairment is a frequent condition in patients with psoriasis vulgaris. The related-purine derivatives may be specific serological markers for identifying those patients with psoriasis vulgaris at a high risk of developing renal dysfunctions.

Acknowledgements
Not applicable.

Funding
No funding was received.

Availability of data and materials
All data generated or analyzed during this study are included in this published article.

Authors’ contributions
IN, MT, CM and SRG conceived the study, identified and reviewed the literature. CIM, MIM and MIS collected the data. CDE and CE analyzed and interpreted the data. All authors equally contributed to writing the manuscript, and CDE and CE edited and revised the manuscript. All authors read and approved the final version of the manuscript.

Ethics approval and consent to participate
The study was approved by the Ethics Committee of ‘Victor Babes’ Clinical Hospital for Infectious Diseases (Bucharest, Romania). All the participants gave their consent to the use of their biological samples in research studies.

Patient consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

References


