Rom J Morphol Embryol 2020, 61(2):503–511 ISSN (print) 1220–0522, ISSN (online) 2066–8279 doi: 10.47162/RJME.61.2.20

ORIGINAL PAPER

RJME Romanian Journal of Morphology & Embryology http://www.rjme.ro/

The role of non-invasive scores in determining the liver fibrosis in NAFLD and psoriatic patients

CARMEN-DANIELA NEAGOE[1)], ANCA-SMARANDA FARMAZON[2)], ANCA-MARIA AMZOLINI[3)], CRISTINA-ELENA SINGER[4)], SIMONA-LAURA IANOŞI[5)], CRISTINA-VIOLETA TUTUNARU[5)], AMELIA-VALENTINA GENUNCHE-DUMITRESCU[1)], NICOLAE-GABRIEL IANOŞI[6)], ION PĂUN[7)], POLLIANA-MIHAELA LERU[8)], OANA-SORINA TICA[9)], MIHAELA POPESCU[10)]

[1)] *1st Department of Internal Medicine, University of Medicine and Pharmacy of Craiova, Romania*

[2)] *PhD Student, Department of Pharmacology, University of Medicine and Pharmacy of Craiova, Romania*

[3)] *2nd Department of Internal Medicine, University of Medicine and Pharmacy of Craiova, Romania*

[4)] *2nd Department of Pediatrics, University of Medicine and Pharmacy of Craiova, Romania*

[5)] *Department of Dermatology, University of Medicine and Pharmacy of Craiova, Romania*

[6)] *Department of Surgery, Dr. Ianoşi Medical Center, Private Practice, Craiova, Romania*

[7)] *Department of Surgery, University of Medicine and Pharmacy of Craiova, Romania*

[8)] *Department of Family Medicine, Carol Davila University of Medicine and Pharmacy, Bucharest, Romania*

[9)] *Department of Pharmacology, University of Medicine and Pharmacy of Craiova, Romania*

[10)] *Department of Endocrinology, University of Medicine and Pharmacy of Craiova, Romania*

Abstract

According to recent data, psoriatic patients have an increased prevalence of non-alcoholic fatty liver disease (NAFLD) and metabolic syndrome, compared with the general population. In some published studies, the severity and presence of psoriasis disease were correlated with the severity of NAFLD. In the current study, we aimed to compare the sensibility and specificity of the non-invasive scores and liver biopsy in determining fibrosis in patients with NAFLD and moderate to severe psoriasis. We performed the scientific research from June 2014–December 2017 and we included 71 patients: 40 patients with NAFLD and 31 patients with moderate to severe psoriasis according to Psoriasis Area and Severity Index (PASI) score and NAFLD, who received Etanercept treatment for at least one year. Based on the clinical and laboratory data, we calculated the following scores for fibrosis: body mass index (BMI), aspartate aminotransferase (AST)/ alanine aminotransferase (ALT) ratio, diabetes (BARD) score, Fibrosis-4 (FIB-4) score, and NAFLD fibrosis score (NFS). For liver biopsy, we used the Menghini technique. By calculating Kendall's test, we also observed a strong direct correlation between the degree of fibrosis and FIB-4 (*tau*=0.558) and NFS (*tau*=0.490) scores, with a critical statistical impact, and the lack of a correlation with the BARD score (*tau*=0.095; *p*=0.332). The hepatic biopsy allowed the more accurate establishment of the role of the non-invasive tests in the diagnosis of the lesions of steatosis, steatohepatitis, and hepatic fibrosis. The non-invasive tests are most useful for the exclusion of the evolution lesions and for the confirmation of the advanced stages of the disease. Among these, the NFS score proved a high statistically significant correlation ($p<0.0001$) with the fibrosis histological lesions.

Keywords: NAFLD, psoriasis, liver biopsy, non-invasive scores, fibrosis.

Introduction

Though apparently unrelated diseases, psoriasis and non-alcoholic fatty liver disease (NAFLD) are linked through many aspects. Although psoriasis disease, a complex and immune-mediated condition, most often affects the skin, it is actually considered a systemic disease associated with multiple extracutaneous systemic manifestations and comorbidities conditions, such as NAFLD, metabolic syndrome, obesity, cardiovascular disease, psoriatic arthritis. The prevalence of psoriasis is 11% in Norway and up to 3% in the US [1, 2].

It has been shown that patients with psoriasis have a higher prevalence of non-alcoholic fatty liver and metabolic syndrome compared to the rest of the population. The latest studies have shown NAFLD was positively correlated with the prevalence and severity of psoriasis. Non-alcoholic fatty liver has been correlated with the prevalence and severity of psoriasis [1, 3–5]. It seems that patients with NAFLD and psoriasis have an increased risk of developing advanced fibrosis compared to patients with NAFLD but without psoriasis [6–8]. In over 80% of cases of psoriasis, the affected body surface area is less than 10%. It seems that patients with NAFLD and psoriasis have an increased risk of developing advanced fibrosis compared to patients with NAFLD but without psoriasis [9]. The possible link between moderate to severe psoriasis, obesity and metabolic syndrome, which are known risk factors for NAFLD, has been recently documented focusing on the important role of the adipose tissue in the development of the inflammatory background shared by the above entities. Adipose tissue acts as an endocrine

organ and has important roles in psoriasis and NAFLD pathogenesis. NAFLD and moderate to severe psoriasis are associated with increased levels of proinflammatory adipokines and decreased levels of anti-inflammatory adipokines, given the common inflammatory etiology. NAFLD is defined as an excessive accumulation of triglycerides in hepatocytes and includes from relatively benign steatosis to non-alcoholic steatohepatitis (NASH), where fatty infiltration is accompanied by inflammation, hepatocellular ballooning, pericellular fibrosis and cirrhosis. NASH occurs in 20% of NAFLD patients and globally has a greater tendency to progress in psoriatic patients [10–12]. The natural history of NAFLD depends on the histological type at the time of the diagnosis, so, if simple steatosis, the progression of the disease might not occur, for some of them, the lesions progress to steatohepatitis, fibrosis and hepatic cirrhosis. The real evaluation of the progression rate of the fibrosis in patients suffering from NASH is limited because studies are retrospective and few patients have had repeated biopsies during the follow-up period [13, 14]. The "golden standard" in the diagnosis of the histological stage of NAFLD is represented by the hepatic biopsy, but it presents plenty of disadvantages and problems in the clinical practice, such as intra- and inter-observational variability, the considerable error rate in the absence of some pathognomonic histological modifications, especially by the lack of a consensus regarding the indications of the biopsy in NAFLD [15], high cost, invasive method, low adherence of the patient, risk of local or systemic complications up to death. According to the different prognosis of the histological types of NAFLD (steatosis, steatohepatitis, fibrosis) and considering that the biopsy involves a lot of risks, some researchers developed numerous noninvasive methods for the management of NAFLD patients knowing that the presence of the hepatic fibrosis is associated with increased risk of hepatic carcinoma and cirrhosis. These methods combine a series of biochemical parameters in systems of scores or use radiological techniques, and try to fulfill the requirements of an ideal non-invasive marker: cost-efficiency, easiness in measuring, hepatic specificity, reproducibility between the patients and different population groups, predictive value [15, 16]. Up to now, there is no single noninvasive method with enough accuracy to allow the replacement of the hepatic biopsy, but if we combine some of them, we get a good sensibility and specificity compared to biopsy, therefore, we can reduce the number of patients who undergo a biopsy to establish the progression of the disease [17].

Aim

In this study, we compared the specificity and sensibility of the noninvasive liver scores with hepatic biopsy in determining the liver fibrosis at NAFLD patients and with psoriasis (moderate to severe).

Patients, Materials and Methods

This study was conducted following the principles of the Helsinki Declaration and Good Clinical Practice and was approved by the Ethics Committee of Dr. Ianoşi Medical Center and the Emergency County Hospital in Craiova, Romania. All patients gave written informed consent. We conducted scientific research in June 2014–December 2017 and initially included 71 patients: 31 patients with psoriasis (moderate or severe), according to the Psoriasis Area and Severity Index (PASI) and NAFLD scores, who received treatment with Etanercept for a period of one year and 40 patients with NAFLD. The inclusion criteria were patients over 18 years of age, with moderate to severe psoriasis and no hepatotoxicity treatment in the last two years. We chose those patients treated with Etanercept, because recent research has shown that Etanercept reduces the risk of fibrosis in patients with psoriasis and NAFLD by lowering the insulin resistance index and improving serum glucose levels. Methotrexate-induced liver histological lesions appear to be similar to those in NAFLD, so we excluded patients on Methotrexate treatment and accepted those who were at least two years old from the last administration and no more than five years of Methotrexate during lifetime.

After anamnestic exclusion of the alcohol consumption or other known hepatic diseases (hemochromatosis, Wilson disease, α_1-antitrypsin deficiency, viral chronic hepatitis), the usage of nonsteroidal anti-inflammatory drugs (NSAIDs) more than two days/week was an exclusion criterion. After signing the written informed consent form for performing the hepatic biopsy, the study group consisted of 50 patients: 18 patients with psoriasis and NAFLD that formed the psoriasis group and 32 patients with NAFLD, forming NAFLD group. In both groups, the subjects were successively enrolled if they met the inclusion criteria. For all 50 patients, we collected: personal data; medical history; anthropometric data [height, weight, body mass index (BMI), abdominal circumference (AC), hip circumference (HC), AC/HC ratio (between the two circumferences)]; biological parameters (blood count, glycemia, total cholesterol and the fractions of cholesterol, triglycerides, insulinemia, transaminases, gamma-glutamyl transferase, albumin); transabdominal ultrasound; histological examination of biopsy samples, and we calculated noninvasive tests for fibrosis. Alcohol consumption was excluded by one minute of the patient's history using a questionnaire – Risk Behavior Monitoring System 2006 and by questioning the relatives, noting the amount of alcohol consumed per day and week and the type of alcohol. We accepted an amount less than 20 g pure alcohol/day for women and less than 30 g pure alcohol/day for men, maximum two times a week. For calculation and interpretation of BMI, we measured the weight and height of patients using a thaliometer with a scale and entered the results obtained in the following formula: BMI (kg/m^2) = current weight (kg)/height^2 (m^2). We measured AC using an inch of tailoring, halfway between the upper iliac crest and the edge of the rib, at the navel, on the median axillary line. Using the waist top, we calculated HC in the gluteal area at the largest diameter.

After measuring HC, we calculated AC/HC ratio (between the two circumferences) to determine the type of adipose tissue distribution: AC/HC $\geq$0.85 for women and $\geq$0.9 for men, for abdominal obesity. We diagnosed the metabolic syndrome on three of the following criteria: AC >80 cm in women and >94 cm in men; serum trigly-

cerides value ≥150 mg/dL (1.7 mmol/L) or lipid reduction treatment; high-density lipoprotein (HDL)-cholesterol in women <50 mg/dL (1.3 mmol/L), in men <40 mg/dL (1 mmol/L), or hypocholesterolemic treatment; blood pressure >130/85 mmHg or hypotensive drugs; blood glucose a young ≥100 mg% or hypoglycemic treatment. The insulin resistance index [homeostatic model assessment of insulin resistance (HOMA-IR)] was calculated using the formula: HOMA-IR index = insulinemia (mU/L) × blood glucose (mmol/L) / 22.5. We considered that HOMA-IR index is pathological at high value of 3. The collection of blood samples for biological explorations was made *à jeun*, from venous blood, after 12 hours of fasting. Using clinical and laboratory data, we calculated fibrosis scores: BMI, aspartate aminotransferase (AST)/ alanine aminotransferase (ALT) ratio, diabetes (BARD) score, Fibrosis-4 (FIB-4) score, and NAFLD fibrosis score (NFS) [18–21]. The BARD score was calculated as the sum of three variables: BMI >28 kg/m^2 – 1 point, AST/ALT ratio >0.8 – 1 point, the presence of diabetes – 1 point. A value less than 2 excludes significant fibrosis [22–25]. According to Brunt's classification, there are five stages: stage 0 means without fibrosis, then in stage 1 we find the characteristic fibrosis pattern in NAFLD, the initial deposit of the perisinusoidal extracellular matrix (area 3). In evolution, in stage 2 occurs periportal fibrosis with formation of fibrous septa, subsequently fibrosis in bridges and, finally, in stage 4 we find cirrhosis [26, 27].

The transabdominal ultrasound was performed by the same doctor on all patients using a Hitachi EUB-8500 with an external convex transducer that transmits a 3.5–5 MHz frequency, representing the used method for the diagnosis of NAFLD that determine if the patients met the criteria or not. Ultrasound criteria that we used to define hepatic steatosis were increased hepato-renal contrast, liver brightness, deep attenuation, and vascular blurring. We considered steatosis if we have got two from four criteria.

For the statistical analysis, the measured parameters were stocked in Excel files, and for the data processing, one used Microsoft Excel (Microsoft Corp., Redmond, WA, USA) program, with the XLSTAT suit for MS Excel (Addinsoft SARL, Paris, France). All the continual variables were reported as average ± standard deviation (SD). For the comparison of the noninvasive scores and results of the histological examination, we used the χ^2 (*chi*-square) test and Kendall's test. The *p*-value <0.05 was considered statistically significant. The Kendall's rank coefficient is often used as a test statistic in a statistical hypothesis test to establish whether two variables may be regarded as statistically dependent. This test is non-parametric. The denominator is the total number of pair combinations, so the coefficient must be in the range $-1 \leq \tau \leq 1$. If the agreement between the two rankings is perfect (*i.e.*, the two rankings are the same), the coefficient has value 1. If the disagreement between the two rankings is perfect (*i.e.*, one ranking is the reverse of the other), the coefficient has value −1. If *X* and *Y* are independent, then we would expect the coefficient to be approximately zero.

Results

In the psoriasis group, we enrolled mostly women compared to men (14:4), unlike the NAFLD group where there were no statistically significant differences (18:14). The mean age in the NAFLD group was 54.25 years and in the psoriasis group was 61.93 years, statistically important difference. Regarding the AC, BMI, metabolic syndrome, the level of cholesterol, there were no major discrepancies between the two study groups. As we expected, HOMA-IR index, triglycerides level and AST level were significantly higher in the psoriasis group. Table 1 presents the biological and clinical characteristics of the patients from the study group.

Table 1 – ***The clinical and biological characteristics of the patients from study group***

	NAFLD group (*n*=32)	Psoriasis group (*n*=18)	*p*-value Student's *t*-test or **chi*-square test
Mean age [years]	54.25± 12.5	61.93± 6.47	0.0034 (S)
Gender (men/women) (*n*)	14/18	4/14	*0.052 (NS–S)
Diabetes mellitus (absent/present) (*n*)	18/14	6/12	*0.058 (NS)
Metabolic syndrome (absent/present) (*n*)	4/28	1/17	*0.486 (NS)
AC [cm]	102.92± 10.72	106± 11.34	0.373 (NS)
AC/HC ratio	0.94± 0.09	0.95± 0.09	0.131 (NS)
BMI [kg/m^2]	30.15± 3.73	31.29± 5.32	0.396 (NS)
Normal weight (*n*)	3	2	
Overweight (*n*)	11	3	0.535 (NS)
1st degree obesity (*n*)	15	10	
2nd degree obesity (*n*)	3	2	
3rd degree obesity (*n*)	0	1	
HOMA-IR index	4.50± 3.96	25.33± 17.4	0.032 (S)
Total cholesterol [mg/dL]	204.72± 41.32	200.93± 42.36	0.773 (NS)
HDL-cholesterol [mg/dL]	48.75± 15.12	39.57± 12.96	0.051 (NS–S)
Triglycerides [mg/dL]	167.68± 84.31	221.71± 70.78	0.039 (S)
AST [IU/L]	40.17± 22.63	54.93± 17.42	0.032 (S)
ALT [IU/L]	53.08± 38.37	64.29± 23.82	0.315 (NS)
AST/ALT ratio	0.85± 0.24	0.95± 0.43	0.321 (NS)
GGT [IU/L]	63.58± 52.62	79.79± 57.23	0.545 (NS)

AC: Abdominal circumference; ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; BMI: Body mass index; GGT: Gamma-glutamyl transferase; HC: Hip circumference; HDL: High-density lipoprotein; HOMA-IR: Homeostatic model assessment of insulin resistance; *n*: No. of patients; NAFLD: Non-alcoholic fatty liver disease; NS: Not significant; S: Significant.

Fibrosis in the studied hepatic samples represented a heterogeneous process. We identified the fibrosis of area 3, perivenular, perisinusoidal/pericellular with focal or extended distribution in 16 cases, which were included in stage F1 in accordance with Brunt's classification and representing the predominant category (Figures 1 and 2). In nine cases, we observed the presence of focal or

extensive periportal fibrosis, the cases being included in the stage (F2) (Figure 3) (Table 2). The presence of fibrosis in bridges was identified in four cases (F3) (Figure 4; Figure 5, a and b), and cirrhosis in a single case (F4) (Figure 6). In Table 3, we show the number of patients according to fibrosis's degree. By comparing the two groups in terms of fibrosis, we can affirm that it is slightly increased in psoriasis group compared to the control group (61.11% *vs.* 59.37%), but if we evaluate the patients in terms of fibrosis degree, we find that the high degree of fibrosis is significantly larger in the psoriasis group (72.7%) than in control group (31.57%). From all the 30 hepatic tissue fragments which presented a degree of fibrosis, in most of the cases (88%), we noticed a more reduced display of the fibrosis on the parenchyma subjacent to the Glisson's capsule, and the most severe fibrosis was noticed in the profound parenchyma in both groups. The clinical and biological characteristics depending on the degree of fibrosis (F1 degree being considered the non-significant fibrosis), are summarized in Table 4. Statistically significant differences were recorded regarding age, AST, albumin and number of platelets between group with advanced fibrosis and those groups with no/mild fibrosis (F0, F1), so that patients with advanced fibrosis were older, with a higher level of AST and a low serum level of albumin and platelet count. The BARD score was not correlated to the degree of fibrosis (p=0.791, *chi*-square test) (Figure 7), FIB-4 score was significantly correlated to the hepatic fibrosis (p=0.0011, *chi*-square test and p=0.004, Fischer's exact test) (Table 5). In our study, we found a statistically highly significant correlation between the degree of fibrosis and the NFS score (p<0.0001, *chi*-square and Fisher's exact tests) (Table 6). By calculating Kendall's test (Table 7), we also observed a strong direct correlation between the fibrosis' degree and FIB-4 (*tau*=0.558) and NFS (*tau*=0.490) scores, with a critical statistical impact, and the lack of a correlation with the BARD score (*tau*=0.095; p=0.332). For all predictive scores of liver fibrosis, the sensitivity values were high, but we found low values of specificity (Table 8). By using the three scores to exclude the significant fibrosis, the hepatic biopsy can be avoided in 48% of the patients by using the FIB-4 score and in 42% of them with the NFS score, but with no false-negative results in the last case (Table 9).

Table 2 – ***The histological features of the patients from study group***

Histological stage	NAFLD group *n* (%)	Psoriasis group *n* (%)
Steatosis	8 (25%)	4 (22.22%)
Inflammation	5 (15.63%)	3 (16.67%)
Fibrosis	19 (59.37%)	10 (55.56%)
Cirrhosis	–	1 (5.55%)
Total	32 (64%)	18 (36%)

n: No. of patients; NAFLD: Non-alcoholic fatty liver disease.

Table 3 – ***The staging of fibrosis in both groups***

Stage of fibrosis	NAFLD group (*n*)	Psoriasis group (*n*)
1A	10	1
1B	2	0
1C	1	2
2	5	4
3	1	3
4	–	1
Total	19	11

n: No. of patients; NAFLD: Non-alcoholic fatty liver disease.

Table 4 – ***The clinical and biological characteristics of the patients depending on the degree of fibrosis***

	Fibrosis F0, F1 stages (*n*=36)	Significant fibrosis F2, F3, F4 stages (*n*=14)	*p*-value Student's *t*-test or **chi*-square test
Mean age [years]	55.15± 12.5	61.12± 5.37	0.004 (S)
Gender (men/women) (*n*)	12/24	6/8	*0.528 (NS)
Diabetes mellitus (absent/present) (*n*)	20/16	4/10	*0.086 (NS)
Metabolic syndrome (absent/present) (*n*)	4/32	1/13	*0.674 (NS)
AC [cm]	103.96± 11	106± 8	0.855 (NS)
AC/HC ratio	0.95± 0.08	0.96± 0.09	0.137 (NS)
BMI [kg/m^2]	30.45± 4.73	31.49± 6.32	0.386 (NS)
Thrombocytes [×10^9/L]	266± 55.46	185.14± 47.19	0.00086 (HS)
Total cholesterol [mg/dL]	204.32± 41.32	200.92± 42.15	0.677 (NS)
HDL-cholesterol [mg/dL]	48.8± 15.1	36.6± 13	0.106 (NS)
Triglycerides [mg/dL]	169± 84	222± 71	0.664 (NS)
Triglycerides/HDL-cholesterol ratio	3.94± 3.14	6.08± 2.38	0.304 (NS)
AST [IU/L]	40.46± 23.11	54.92± 17.14	<0.0001 (HS)
ALT [IU/L]	53.18± 37	64.28± 24	0.054 (NS)
AST/ALT ratio	0.84± 0.26	0.94± 0.44	0.331 (NS)
GGT [IU/L]	63.68± 42.62	79.88± 47.22	0.534 (NS)
Albumin [g/dL]	4.22± 0.3	2.97± 0.35	0.00041 (HS)
FIB-4 score	1.24± 0.63	2.74± 1.99	<0.0001 (HS)
BARD score	2.3 (0–4)	2.71 (1–4)	0.153 (NS)
NFS score	-1.73± 1.43	0.28± 1.08	0.00086 (HS)

AC: Abdominal circumference; ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; BARD: BMI, AST/ALT ratio, diabetes; BMI: Body mass index; FIB-4: Fibrosis-4; GGT: Gamma-glutamyl transferase; HC: Hip circumference; HDL: High-density lipoprotein; HS: Highly significant; *n*: No. of patients; NAFLD: Non-alcoholic fatty liver disease; NFS: NAFLD fibrosis score; NS: Not significant; S: Significant.

Table 5 – ***The distribution of the patients depending on the fibrosis degree and the FIB-4 score***

FIB-4 score	F0, F1 stages	F2, F3, F4 stages	Total
<1.3	23	1	24
1.3–3.25	12	11	23
>3.25	1	2	3
Total	36	14	50

FIB-4: Fibrosis-4.

Table 6 – ***The distribution of the patients depending on the fibrosis degree and NFS score***

NFS score limits	F0, F1 stages	F2, F3, F4 stages	Total
<-1.455	21	0	21
-1.455–0.676	15	9	24
>0.676	0	5	5
Total	36	14	50

NAFLD: Non-alcoholic fatty liver disease; NFS: NAFLD fibrosis score.

Table 7 – ***Correlations between fibrosis degree and fibrosis scores***

Fibrosis	BARD score	FIB-4 score	NFS score
Kendall's *tau*	0.095	0.558	0.490
Kendall's *p*-value	0.332	<0.0001	<0.0001

ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; BARD: BMI, AST/ALT ratio, diabetes; BMI: Body mass index; FIB-4: Fibrosis-4; NAFLD: Non-alcoholic fatty liver disease; NFS: NAFLD fibrosis score.

Table 8 – ***The sensibility and specificity of the scores in detection of significant fibrosis***

F2, F3, F4 stages	BARD score >2	FIB-4 score >1.3	NFS score >-1.455
Sensitivity	78.57%	92.86%	100.00%
Specificity	25.00%	63.89%	58.33%

ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; BARD: BMI, AST/ALT ratio, diabetes; BMI: Body mass index; FIB-4: Fibrosis-4; NAFLD: Non-alcoholic fatty liver disease; NFS: NAFLD fibrosis score.

Table 9 – ***The proportion of the patients who can avoid the hepatic biopsy by using non-invasive tests for the exclusion of the significant fibrosis***

Score	Cut-off	*n* (%)	False negative results, *n* (%)
BARD	<2	12/50 (24%)	3 (25%)
FIB-4	<1.3	24/50 (48%)	1 (4%)
NFS	<-1.455	22/50 (42%)	0 (0%)

ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; BARD: BMI, AST/ALT ratio, diabetes; BMI: Body mass index; FIB-4: Fibrosis-4; *n*: No. of patients; NAFLD: Non-alcoholic fatty liver disease; NFS: NAFLD fibrosis score.

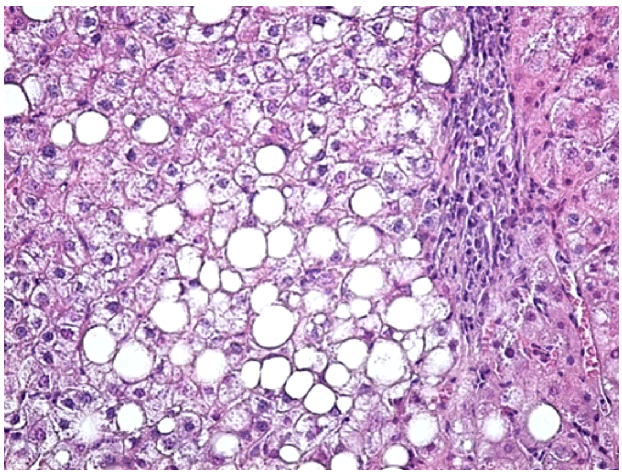

Figure 1 – ***NAFLD F1 stage: macro- and microvesicular steatosis, hepatocyte ballooning degeneration, portal space fibrosis and lymphoplasmocytic inflammatory infiltrate (HE staining, ×200). NAFLD: Non-alcoholic fatty liver disease.***

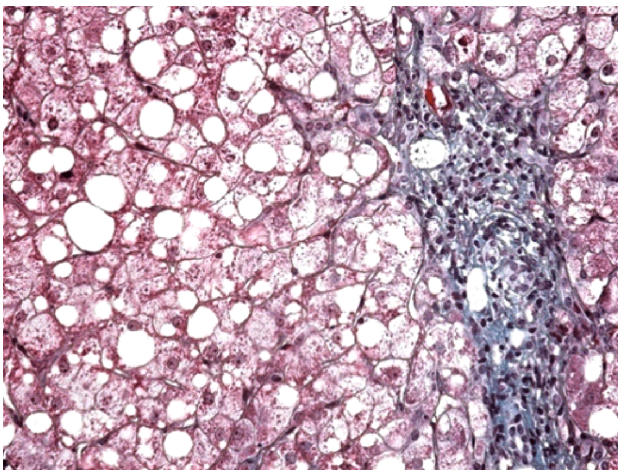

Figure 2 – ***NAFLD F1 stage: macro- and microvesicular steatosis, hepatocyte ballooning degeneration, portal space fibrosis and lymphoplasmocytic inflammatory infiltrate (Masson's trichrome staining, ×100). NAFLD: Non-alcoholic fatty liver disease.***

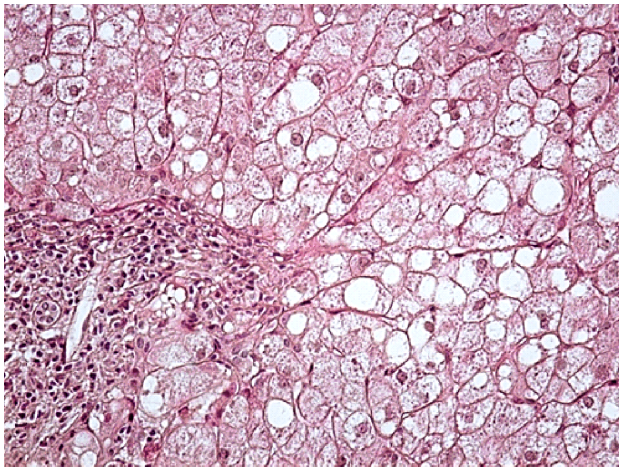

Figure 3 – ***NAFLD F2 stage: macro- and microvesicular steatosis, hepatocyte ballooning degeneration, perisinusoidal fibrosis, portal space lymphoplasmocytic inflammatory infiltrate (HE staining, ×100). NAFLD: Non-alcoholic fatty liver disease.***

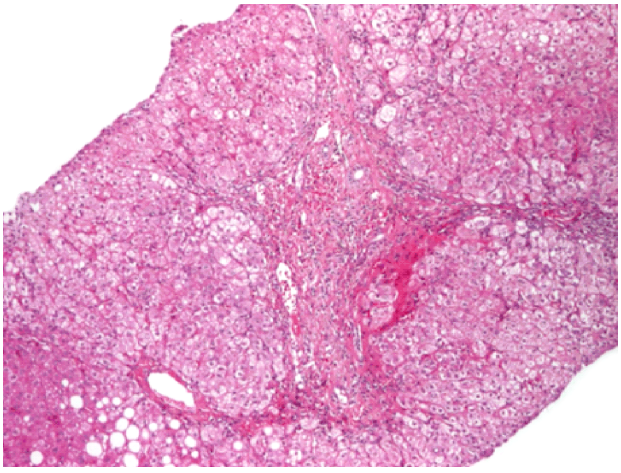

Figure 4 – ***NAFLD F3 stage: bridging fibrosis (HE staining, ×40). NAFLD: Non-alcoholic fatty liver disease.***

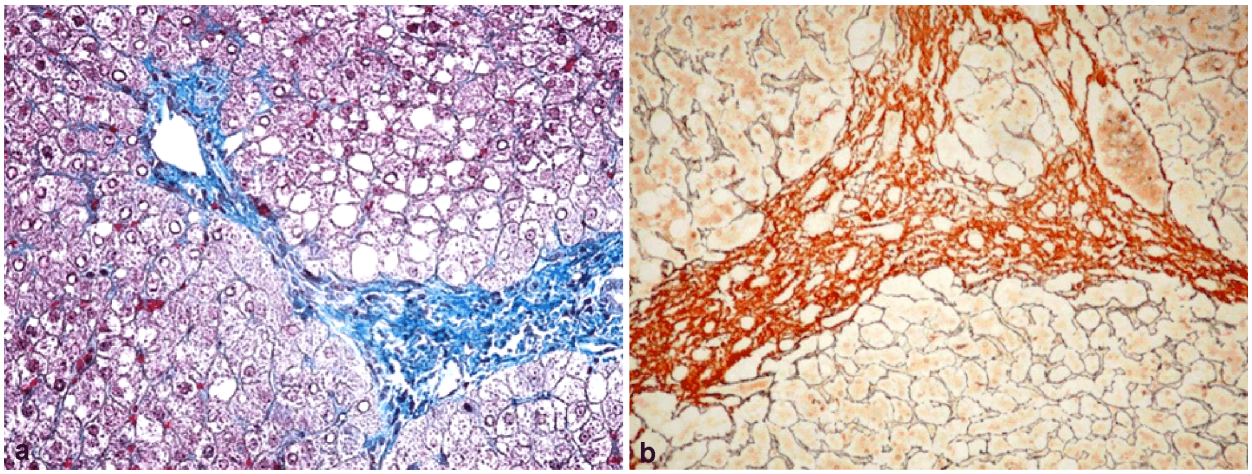

Figure 5 – ***(a) Steatohepatitis F3 stage: porto-central fibrous septa, hepatocyte degeneration and steatosis (Masson's trichrome staining, ×100); (b) Bridging fibrosis F4 stage (Argentic staining, ×100).***

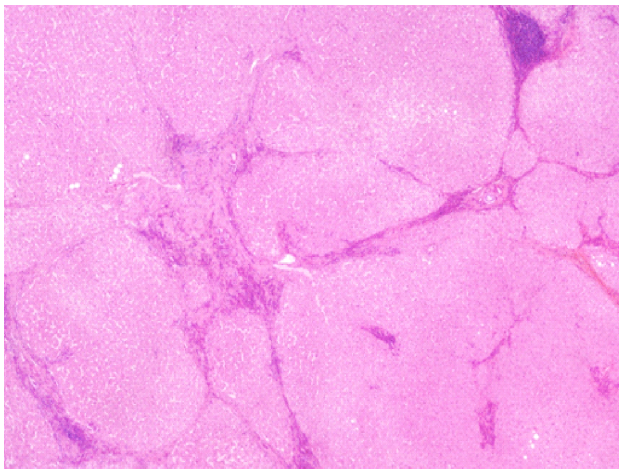

Figure 6 – ***Macronodular cirrhosis. HE staining, ×40.***

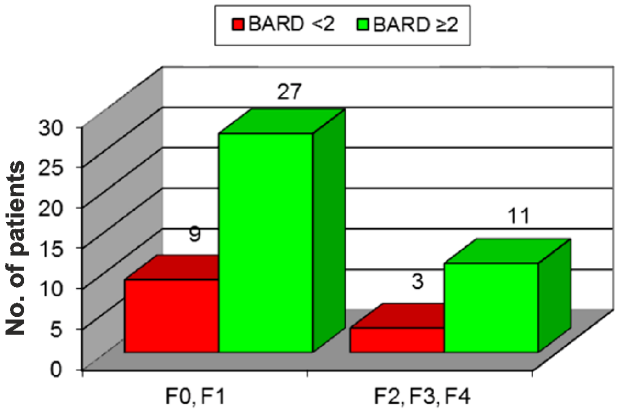

Figure 7 – ***The distribution of patients depending on the BARD score and the fibrosis (F) degree. ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; BARD: BMI, AST/ALT ratio, diabetes; BMI: Body mass index.***

Discussions

Various studies have shown that NAFLD prevalence is higher among patients with psoriasis. To understand this, we must observe the similarities between the pathogenesis of psoriasis disease and NAFLD. Lonardo *et al.* described for the first time in 2001, three simultaneous cases of psoriasis and NASH (diagnosed by biopsy), then in 2004, Matsumoto *et al.* presented the case of a young obese psoriatic patient diagnosed with NASH, whose histopathological appearance improved after a period of hypocaloric diet. Two important studies regarding the link between NAFLD and psoriasis were made by Gisondi *et al.* and Miele *et al.* have shown that the occurrence of NAFLD is higher among patients with psoriasis (47% *versus* 28%) and has been correlated with the psoriasis severity assessed by steps. In Miele *et al.* study, the NAFLD occurrence was 59.2% of a group of 142 patients but there has been found no correlation with the severity of psoriasis [28–31]. The liver biopsy, although it is an invasive procedure difficult to accept by patients, it remains the "gold standard" in diagnosing and staging of NAFLD. Therefore, it is important to develop mathematical scores with high sensitivity and specificity, which will allow doctors to better assess liver damage. The serological FibroMax test for assessment of steatosis, inflammation, and fibrosis, as well as different elastography techniques are nowadays accepted, but neither one can be routinely used in the general practitioner or dermatologist office. Etiological factors involved in growth prevalence of NAFLD in those with psoriasis are little known.

The insulin target tissues are the liver, the adipose tissue and skeletal muscle. In the liver, the insulin regulates glucose metabolism, while in the adipose tissue, insulin reduces the hormones' sensitive lipase activity leading to the prevention of free fatty acids output from adipocytes. In the adipose tissue, insulin esterifies free fatty acids and the storage of triglycerides. When installing insulin resistance, free fatty acids are stored at the hepatocyte level, their accumulation leading to lipotoxicity. In this study, the calculated value of HOMA-IR index was higher among patients suffering from psoriasis compared to the NAFLD group, and the increasing insulin resistance

contributes to the emergence of the NAFLD group. Besides, patients with advanced fibrosis had a value of HOMA-IR index compared to patients with low fibrosis (F0, F1).

Many studies have shown that an increase in insulin resistance is common among psoriatic patients and proves that insulin resistance can contribute to the emergence of NAFLD through a variety of mechanisms. Current studies have shown that tumor necrosis factor-alpha (TNF-α) produced by the liver in response to the accumulation of fatty acids determine an increased resistance, therefore insulin resistance is responsible for the worsening of skin lesions in psoriasis. The most common histological changes in psoriasis are proliferation, abnormal keratinocyte differentiation, angiogenesis, and activated cluster of differentiation (CD) 4, CD8, T-lymphocytes in the dermis and epidermis.

Natural killer (NK) cells initiate the inflammatory process through proinflammatory cytokines [32]. Although 80% of patients with psoriasis have the disease limited to <10% of the body surface, psoriasis is not a life-threatening disease, however, the quality of life decreases a lot in these patients [33, 34]. Proinflammatory cytokines like TNF-α and interleukin-6 (IL-6) determine metabolic disorders in psoriasis. Recent studies show that NAFLD represent an important risk factor for cardiovascular events [35]. The patients with psoriasis have low level of HDL-cholesterol and high levels of triglycerides, cholesterol, low-density lipoprotein (LDL)-cholesterol. This dyslipidemic profile leads to the occurrence of psoriasis [36]. In our study, the triglycerides level has been found statistically significantly higher in patients with psoriasis, compared to the control group, but the degree of liver fibrosis could not be correlated with the triglycerides level. Regarding HDL-cholesterol, although we could not establish a statistically significant correlation between the two groups, we could observe that the mean HDL-cholesterol value was normal in patients with NAFLD and slightly below normal in patients with psoriasis. In this research, we did not found a correlation between liver fibrosis and HDL-cholesterol level. Both groups have had sensibly equal cholesterol levels that did not correspond with the degree of liver fibrosis.

Patients with psoriasis and NAFLD have the transaminase (AST/ALT) ratio higher and fibrosis scores higher compared to NAFLD patients without psoriasis. The transaminase ratio did not correlate positively with the degree of hepatic fibrosis in patients with NAFLD. We did not find a correspondence between the transminase ratio, the presence or absence of psoriasis nor with the degree of liver fibrosis. Elevated transaminases are not directly proportional to the degree of liver damage (appreciated through biopsy). Increased inflammation, steatosis and fibrosis was associated with the presence of psoriasis. The small number of patients included in this study does not allow us to draw a firm conclusion. As we have shown in our study, the association between psoriasis and NAFLD increases the risk of worsening liver disease, these patients requiring careful monitoring in order to diagnose and stage the NAFLD early on. On the other hand, we must consider that the patients with psoriasis receive long-term treatment with medication that can potentially harm the liver, further on increasing the risk for hepatic disease. Although the serological determinations for the calculation of these scores are accessible on a large scale, the non-invasive tests are most useful for the exclusion of the evolution lesions and for the confirmation of the advanced stages of the disease. Among these, NFS score proved a high statistically significant correlation with the fibrosis histological lesions.

The importance of our study consists in comparing non-invasive scores with histopathological appearance for both psoriatic patients and control group. There are numerous studies that have evaluated the prevalence of NAFLD in patients with psoriasis or tracked the evolution of liver fibrosis in patients with psoriasis, but most of them have diagnosed NAFLD and fibrosis using ultrasonography and transient elastography (FibroScan). Only in two studies, Miele *et al.* (2009) [28] and Roberts *et al.* (2015) [37], the diagnosis of NAFLD was supported by biopsy. In Miele *et al.* study, only five biopsies were performed in patients with psoriasis, while in Roberts *et al.* study, the number of patients who underwent liver biopsy was 52. Although the number of patients in our study was not large, all patients underwent liver biopsy and the comparison of non-invasive scores was made with histopathological examination. Therefore, we believe that this study is important in validating non-invasive liver fibrosis scores.

Conclusions

As we have shown in our study, the association between psoriasis and NAFLD increases the risk for unfavorable evolution of the hepatic disease, therefore these patients need to be regularly monitored in order to diagnose and stage the NAFLD early on. On the other hand, we must consider that the patients with psoriasis receive long-term treatment with medication that can potentially harm the liver, further on increasing the risk for hepatic disease. The performance of the hepatic biopsy in part of the patients, allowed the more accurate establishment of the role of the non-invasive tests in the diagnosis of the lesions of steatosis, steatohepatitis and hepatic fibrosis. Although the serological determinations for the calculation of these scores are accessible on a large scale, the non-invasive tests are most useful for the exclusion of the evolution lesions and for the confirmation of the advanced stages of the disease. Among these, NFS score proved a high statistically significant correlation with the fibrosis histological lesions.

Conflict of interests

The authors declare that they have no conflict of interests.

Authors' contribution

Carmen-Daniela Neagoe & Anca-Smaranda Farmazon equally contributed to the manuscript and share main authorship.

References

[1] Prussick RB, Miele L. Nonalcoholic fatty liver disease in patients with psoriasis: a consequence of systemic inflammatory burden? Br J Dermatol, 2018, 179(1):16–29. https://doi.org/10.1111/bjd.16239 PMID: 29235656

[2] Rachakonda TD, Schupp CW, Armstrong AW. Psoriasis prevalence among adults in the United States. J Am Acad Dermatol, 2014, 70(3):512–516. https://doi.org/10.1016/j.jaad.2013.11.013 PMID: 24388724

[3] Gisondi P, Targher G, Zoppini G, Girolomoni G. Non-alcoholic fatty liver disease in patients with chronic plaque psoriasis. J Hepatol, 2009, 51(4):758–764. https://doi.org/10.1016/j.jhep.2009.04.020 PMID: 19560226

[4] Madanagobalane S, Anandan S. The increased prevalence of nonalcoholic fatty liver disease in psoriatic patients: a study from South India. Australas J Dermatol, 2012, 53(3):190–197. https://doi.org/10.1111/j.1440-0960.2012.00905.x PMID: 22672067

[5] Abedini R, Salehi M, Lajevardi V, Beygi S. Patients with psoriasis are at a higher risk of developing nonalcoholic fatty liver disease. Clin Exp Dermatol, 2015, 40(7):722–727. https://doi.org/10.1111/ced.12672 PMID: 25958919

[6] Candia R, Ruiz A, Torres-Robles R, Chávez-Tapia N, Méndez-Sánchez N, Arrese M. Risk of non-alcoholic fatty liver disease in patients with psoriasis: a systematic review and meta-analysis. J Eur Acad Dermatol Venereol, 2015, 29(4):656–662. https://doi.org/10.1111/jdv.12847 PMID: 25418531

[7] Al-Mutairi N, Al-Farag S, Al-Mutairi A, Al-Shiltawy M. Comorbidities associated with psoriasis: an experience from the Middle East. J Dermatol, 2010, 37(2):146–155. https://doi.org/10.1111/j.1346-8138.2009.00777.x PMID: 20175849

[8] Yang YW, Keller JJ, Lin HC. Medical comorbidity associated with psoriasis in adults: a population-based study. Br J Dermatol, 2011, 165(5):1037–1043. https://doi.org/10.1111/j.1365-2133.2011.10494.x PMID: 21711339

[9] Prussick R, Prussick L, Nussbaum D. Nonalcoholic fatty liver disease and psoriasis: what a dermatologist needs to know. J Clin Aesthet Dermatol, 2015, 8(3):43–45. PMID: 25852814 PMCID: PMC4382145

[10] Adams LA, Angulo P, Lindor KD. Nonalcoholic fatty liver disease. CMAJ, 2005, 172(7):899–905. https://doi.org/10.1503/cmaj.045232 PMID: 15795412 PMCID: PMC554876

[11] Ganzetti G, Campanati A, Offidani A. Non-alcoholic fatty liver disease and psoriasis: so far, so near. World J Hepatol, 2015, 7(3):315–326. https://doi.org/10.4254/wjh.v7.i3.315 PMID: 25848461 PMCID: PMC4381160

[12] Wenk KS, Arrington KC, Ehrlich A. Psoriasis and non-alcoholic fatty liver disease. J Eur Acad Dermatol Venereol, 2011, 25(4):383–391. https://doi.org/10.1111/j.1468-3083.2010.03841.x PMID: 20840346

[13] Popescu M, Popescu IAS, Stanciu M, Cazacu SM, Ianoşi NG, Comănescu MV, Singer CE, Neagoe CD. Non-alcoholic fatty liver disease – clinical and histopathological aspects. Rom J Morphol Embryol, 2016, 57(4):1295–1302. PMID: 28174796

[14] Fiore M, Leone S, Maraolo AE, Berti E, Damiani G. Liver illness and psoriatic patients. BioMed Res Int, 2018, 2018:3140983. https://doi.org/10.1155/2018/3140983 PMID: 29546055 PMCID: PMC5818942

[15] Chalasani N, Younossi Z, Lavine JE, Charlton M, Cusi K, Rinella M, Harrison SA, Brunt EM, Sanyal AJ. The diagnosis and management of nonalcoholic fatty liver disease: practice guidance from the American Association for the Study of Liver Diseases. Hepatology, 2018, 67(1):328–357. https://doi.org/10.1002/hep.29367 PMID: 28714183

[16] Puri P, Sanyal AJ. Nonalcoholic fatty liver disease: definitions, risk factors, and workup. Clin Liver Dis (Hoboken), 2012, 1(4):99–103. https://doi.org/10.1002/cld.81 PMID: 31186860 PMCID: PMC6499283

[17] Fekete GL, Fekete JE. *Steatocystoma multiplex generalisata partial suppurativa* – case report. Acta Dermatovenereol Croat, 2010, 18(2):114–119. PMID: 20624362

[18] Sun W, Cui H, Li N, Wei Y, Lai S, Yang Y, Yin X, Chen DF. Comparison of FIB-4 index, NAFLD fibrosis score and BARD score for prediction of advanced fibrosis in adult patients with non-alcoholic fatty liver disease: a meta-analysis study. Hepatol Res, 2016, 46(9):862–870. https://doi.org/10.1111/hepr.12647 PMID: 26763834

[19] Guha IN, Parker J, Roderick PR, Harris S, Rosenberg WM. Non-invasive markers associated with liver fibrosis in non-alcoholic fatty liver disease. Gut, 2006, 55(11):1650–1660. https://doi.org/10.1136/gut.2006.091454 PMID: 17047111 PMCID: PMC1860097

[20] Harrison SA, Oliver D, Arnold HL, Gogia S, Neuschwander-Tetri BA. Development and validation of a simple NAFLD clinical scoring system for identifying patients without advanced disease. Gut, 2008, 57(10):1441–1447. https://doi.org/10.1136/gut.2007.146019 PMID: 18390575

[21] Angulo P, Hui JM, Marchesini G, Bugianesi E, George J, Farrell GC, Enders F, Saksena S, Burt AD, Bida JP, Lindor K, Sanderson SO, Lenzi M, Adams LA, Kench J, Therneau TM, Day CP. The NAFLD fibrosis score: a noninvasive system that identifies liver fibrosis in patients with NAFLD. Hepatology, 2007, 45(4):846–854. https://doi.org/10.1002/hep.21496 PMID: 17393509

[22] Lazo M, Clark JM. The epidemiology of nonalcoholic fatty liver disease: a global perspective. Semin Liver Dis, 2008, 28(4):339–350. https://doi.org/10.1055/s-0028-1091978 PMID: 18956290

[23] Musso G, Gambino R, Cassader M, Pagano G. Meta-analysis: natural history of non-alcoholic fatty liver disease (NAFLD) and diagnostic accuracy of non-invasive tests for liver disease severity. Ann Med, 2011, 43(8):617–649. https://doi.org/10.3109/07853890.2010.518623 PMID: 21039302

[24] Wong VWS, Wong GLH, Chim AML, Tse AML, Tsang SWC, Hui AY, Choi PCL, Chan AWH, So WY, Chan FKL, Sung JJY, Chan HLY. Validation of the NAFLD fibrosis score in a Chinese population with low prevalence of advanced fibrosis. Am J Gastroenterol, 2008, 103(7):1682–1688. https://doi.org/10.1111/j.1572-0241.2008.01933.x PMID: 18616651

[25] Kleiner DE, Brunt EM, Van Natta M, Behling C, Contos MJ, Cummings OW, Ferrell LD, Liu YC, Torbenson MS, Unalp-Arida A, Yeh M, McCullough AJ, Sanyal AJ; Nonalcoholic Steatohepatitis Clinical Research Network. Design and validation of histological scoring system for non-alcoholic fatty liver disease. Hepatology, 2005, 41(6):1313–1321. https://doi.org/10.1002/hep.20701 PMID: 15915461

[26] Brunt EM, Janney CG, DiBisceglie AM, Neuschwander-Tetri BA, Bacon BR. Nonalcoholic steatohepatitis: a proposal for grading and staging the histological lesions. Am J Gastroenterol, 1999, 94(9):2467–2474. https://doi.org/10.1111/j.1572-0241.1999.01377.x PMID: 10484010

[27] Brunt EM, Kleinert DE, Wilson LA, Belt P, Neuschwander-Tetri BA; NASH Clinical Research Network (CRN). Non-alcoholic fatty liver disease (NAFLD) activity score and the histopathologic diagnosis in NAFLD: distinct clinicopathologic meanings. Hepatology, 2011, 53(3):810–820. https://doi.org/10.1002/hep.24127 PMID: 21319198 PMCID: PMC3079483

[28] Miele L, Vallone S, Cefalo C, La Torre G, Di Stasi C, Vecchio FM, D'Agostino M, Gabrieli ML, Vero V, Biolato M, Pompili M, Gasbarrini G, Rapaccini G, Amerio P, De Simone C, Grieco A. Prevalence, characteristics and severity of non-alcoholic fatty liver disease in patients with chronic plaque psoriasis. J Hepatol, 2009, 51(4):778–786. https://doi.org/10.1016/j.jhep.2009.06.008 PMID: 19664838

[29] Gisondi P, Tessari G, Conti A, Piaserico S, Schianchi S, Peserico A, Giannetti A, Girolomoni G. Prevalence of metabolic syndrome in patients with psoriasis: a hospital-based case-control study. Br J Dermatol, 2007, 157(1):68–73. https://doi.org/10.1111/j.1365-2133.2007.07986.x PMID: 17553036

[30] Batani A, Brănișteanu DE, Ilie MA, Boda D, Ianosi S, Ianosi G, Caruntu C. Assessment of dermal papillary and microvascular parameters in *psoriasis vulgaris* using *in vivo* reflectance confocal microscopy. Exp Ther Med, 2018, 15(2):1241–1246. https://doi.org/10.3892/etm.2017.5542 PMID: 29434710 PMCID: PMC5774437

[31] Leru PM. Drug allergies in primary care practice in Romania: a questionnaire-based survey. Allergy Asthma Clin Immunol, 2014, 10(1):16. https://doi.org/10.1186/1710-1492-10-16 PMID: 24690448 PMCID: PMC4021609

[32] Ianoşi SL, Forsea AM, Lupu M, Ilie MA, Zurac S, Boda D, Ianosi G, Neagoe D, Tutunaru C, Popa CM, Caruntu C. Role of modern imaging techniques for the *in vivo* diagnosis of lichen planus. Exp Ther Med, 2018, 17(2):1052–1060. https://doi.org/10.3892/etm.2018.6974 PMID: 30679973 PMCID: PMC6327670

[33] Ji J, Shu X, Sundquist K, Sundquist J, Hemminki K. Cancer risk in hospitalised psoriasis patients: a follow-up study in Sweden. Br J Cancer, 2009, 100(9):1499–1502. https://doi.org/10.1038/sj.bjc.6605027 PMID: 19352386 PMCID: PMC2694437

[34] Iordache AM, Docea AO, Buga AM, Mitrut R, Albulescu D, Zlatian O, Ianosi S, Ianosi G, Neagoe D, Sifaki M, Rogoveanu OC, Branisteanu DE, Calina D. The incidence of skin lesions in contrast media-induced chemical hypersensitivity. Exp Ther Med, 2019, 17(2):1113–1124. https://doi.org/10.3892/etm.2018.7056 PMID: 30679982 PMCID: PMC6327547

[35] Czernichow S, Kengne AP, Huxley RR, Batty GD, de Galan B, Grobbee D, Pillai A, Zoungas S, Marre M, Woodward M, Neal B, Chalmers J; ADVANCE Collaborative Group. Comparison of waist-to-hip ratio and other obesity indices as predictors of cardiovascular disease risk in people with type-2 diabetes: a prospective cohort study from ADVANCE. Eur J Cardiovasc Prev Rehabil, 2011, 18(2):312–319. https://doi.org/10.1097/HJR.0b013e32833c1aa3 PMID: 20628304 PMCID: PMC4170784

[36] Kahn HS. The "lipid accumulation product" performs better than the body mass index for recognizing cardiovascular risk: a population-based comparison. BMC Cardiovasc Disord, 2005, 5:26. https://doi.org/10.1186/1471-2261-5-26 PMID: 16150143 PMCID: PMC1236917

[37] Mantovani A, Gisondi P, Lonardo A, Targher G. Relationship between non-alcoholic fatty liver disease and psoriasis: a novel hepato-dermal axis? Int J Mol Sci, 2016, 17(2):217. https://doi.org/10.3390/ijms17020217 PMID: 26861300 PMCID: PMC4783949

Corresponding authors

Simona-Laura Ianoşi, Associate Professor, MD, PhD, Department of Dermatology, University of Medicine and Pharmacy of Craiova, 2 Petru Rareş Street, 200349 Craiova, Dolj County, Romania; Phone +40724–350 004, e-mail: simonaianosi@hotmail.com

Ion Păun, Associate Professor, MD, PhD, Department of Surgery, University of Medicine and Pharmacy of Craiova, 2 Petru Rareş Street, 200349 Craiova, Dolj County, Romania; Phone +40723–347 572, e-mail: dripaun@gmail.com

Received: June 4, 2020

Accepted: September 17, 2020