UNIVERSITY OF MEDICINE AND PHARMACY „CAROL DAVILA”, BUCHAREST

DOCTORAL SCHOOL

MEDICINE

THE IMPACT OF SLEEP DISORDERS AND NEUROCOGNITIVE DYSFUNCTIONS ON THE EVOLUTION OF PATIENTS WITH CHRONIC LIVER DISEASE

PHD THESIS SUMMARY

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Introduction

Chronic liver diseases represent global public health problems, while epidemiological studies show that their incidence and prevalence are constantly increasing.

Hepatology - a vast, intensely and long-studied and at the same time inexhausted subject - captured my interest because of the complexity of the pathologies affecting the entire human body, some of them through incompletely elucidated mechanisms.

In current practice, the focus is set on preventing and treating complications of chronic liver disease in order to increase survival. However, less importance is given to other conditions that may occur during the course of the disease, such as: sleep disorders, neurocognitive dysfunctions (e.g., difficulties in concentration and attention, memory loss, etc.), fatigue, depression, etc., which are often underdiagnosed due to the lack of standardized management.

Starting from this premise, we issued the following **working hypothesis**: there are pathologies in subclinical and inapparent forms, related to chronic liver diseases, which can have a negative impact on patients, starting from early stages, and by associating with risk factors, with other comorbidities and with complications related to liver diseases.

Consequently, I developed the doctoral thesis entitled *"The impact of sleep disorders and neurocognitive dysfunctions on the evolution of patients with chronic liver diseases"* by establishing the following **main objective of the research**: the identification of sleep disorders and neurocognitive dysfunctions that stand for the diagnosis of minimal hepatic encephalopathy (MHE) among patients with chronic liver diseases, begging with precirrhotic stages (simple steatosis, steatohepatitis, chronic hepatitis) and continuing with stages of compensated or decompensated liver cirrhosis.

The thesis includes a general, theoretical part and a special part, with personal contributions.

The general part is structured in three chapters that address the current state of knowledge, comprising a review of the literature in this specific domain.

Chapter 1 provides data on the epidemiology, etiology and management of the most common causes of chronic liver disease.

Chapter 2 is dedicated to the presentation of sleep disorders, with emphasis on the particularities encountered in patients with chronic liver diseases.

Chapter 3 presents general information related to the classification, epidemiology, pathophysiology, clinical manifestations, diagnosis and treatment of hepatic encephalopathy, paying special attention to subclinical neurocognitive dysfunctions in minimal hepatic encephalopathy.

The section regarding personal contributions is organized in chapters 4-9, while chapter 10 presents the research conclusions, own contributions and future perspectives. References and appendices are added at the end of the thesis in a separate section.

Personal contributions

Chapter 4. Working hypothesis and general research objectives

The aforementioned *working hypothesis* was based on the following premises:

- among extrahepatic pathologies, sleep disorders are highly prevalent in patients with chronic liver disease; these manifestations are clinically inapparent and therefore neglected.
- subclinical neurocognitive dysfunctions might represent a social and safety problem for both patients and people around them, being underdiagnosed and therefore untreated.
- the presence of minimal hepatic encephalopathy is imperceptible and its progression to overt hepatic encephalopathy has major clinical and economic implications, contributing to deterioration in quality of life and a significant number of deaths.
- it is well-known that patients with advanced liver disease have a profoundly impaired quality of life due to the presence of liver complications requiring repeated hospitalizations; however, extrahepatic pathologies require further attention as they may also be implicated in worsening liver disease and reducing quality of life, not only in decompensated stages of cirrhosis, but also in compensated or even precirrhotic stages.

The objectives of this research are the following:

a) Evaluation of demographic, anamnestic, clinical and paraclinical characteristics of patients with chronic liver diseases through descriptive and comparative statistical analysis.
b) Identification of associated factors, predictors and correlations with statistical significance between sleep disorders, respectively neurocognitive dysfunctions in MHE and risk

behaviors, comorbidities, demographic, anamnestic, clinical and paraclinical parameters of the enrolled patients.

c) Reassessment of sleep disturbances and neurocognitive dysfunctions at 6-month interval by appreciating their impact on patients' outcome.

Chapter 5. General research methodology

The present research is a prospective, observational, multidisciplinary study, which enrolled 178 patients, in accordance with the ethical standards of the Helsinki Declaration of 1975 (revised in 2008), with the approval of the ethics committee of the Clinical Emergency Hospital of Bucharest and the informed consent of the patients. Both outpatients and inpatients were included. For inpatients, clinical and paraclinical parameters were recorded either on the day of discharge or 1-2 days after discharge.

The inclusion criteria included:

- adults aged ≥ 18 years, conscious, cooperative, who provided verbal and written consent for inclusion in the study;
- chronic liver disease defined by steatosis, steatohepatitis, chronic hepatitis, liver cirrhosis, according to clinical, paraclinical - laboratory, upper digestive endoscopy, imaging (CT, MRI) and ultrasound (including ARFI / Acoustic Radiation Force Impulse using ultrasound model Siemens Acuson S2000TM, available in the Digestive Endoscopy Laboratory of the Clinical Emergency Hospital of Bucharest).

The exclusion criteria included:

- patients who refused to sign informed consent;
- patients who are unable to write/read/ have hearing, speech and/or uncorrected vision impairments;
- life-threatening conditions such as acute liver failure, haemodynamic instability, coma, acute myocardial infarction, stroke, advanced decompensated cirrhosis with imminent risk of death, alcohol withdrawal syndrome.
- acute hepatitis, regardless of etiology (e.g., alcoholic, drug-induced, viral).
- overt hepatic encephalopathy, at the time of enrolment (stages II, III and IV according to WEST-HAVEN classification).
- chronic liver disease of unknown/cryptogenic etiology.

- patients whose short and medium-term prognosis was assessed as being unfavorable (including those with hepatic or extrahepatic malignancies).
- history of neurological and/or psychiatric disorders and/or sleep disorders, with/without specific treatment.
- acute renal failure/ creatinine > 2 mg/dl, uncorrected, at the time of study enrollment.
- patients undergoing renal dialysis.
- patients with recent ethanol consumption (>30g/day for men, >20g/day for women) in the last 2 weeks prior to study enrollment.
- patients working in night shifts.

Patients were divided into *2 main groups*:

- group 1– included 96 patients with chronic liver disease in pre-cirrhotic stages* (steatosis, steatohepatitis, chronic hepatitis known for at least 6 months), without clinical, paraclinical and/or imaging, ultrasound and/or endoscopic signs; * for simplification, throughout the research, we used the term "pre- cirrhotic "/" pre- cirrhotic patients", referring to this group of patients.
- group 2 – 82 patients with cirrhosis diagnosed by clinical, paraclinical, imaging/ultrasound/endoscopic parameters and subclassified according to Child-Pugh score into 2 subgroups: compensated cirrhosis (Child A) and decompensated cirrhosis (Child B and C).

Demographic, anamnestic, clinical and paraclinical data were collected from all patients and analyzed in Chapter 6. Patients were administered the psychometric test battery (Chapter 7) and asked to complete sleep questionnaires (Chapter 8). Of the 178 patients enrolled, 104 patients were also assessed using actigraphy/actiwatch (Chapter 8). Out of the 104 patients, 48 patients were reassessed after 6 months.

The distribution of patients regarding groups division and methodology can be seen below (Figure 1).

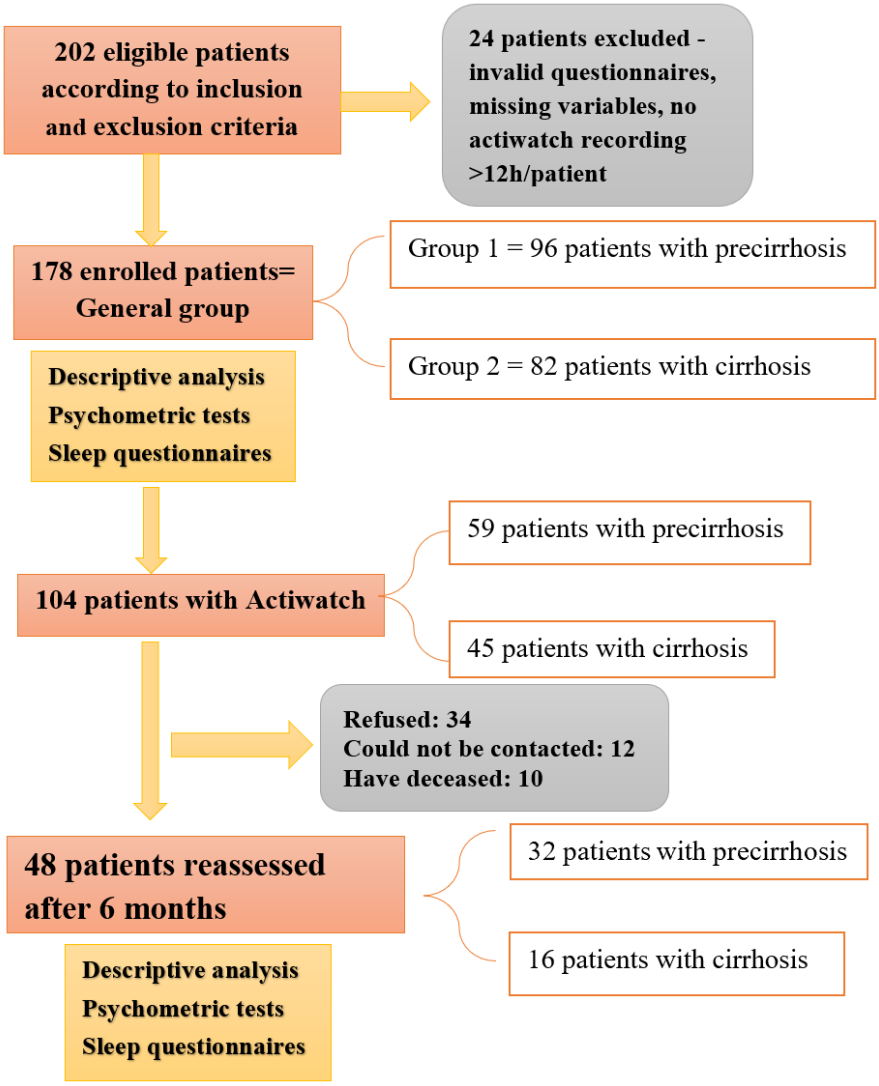

Figura 1. Flow-chart regarding study methodology and groups division

Statistical data analysis. The data were collected and processed with the Microsoft Office 2016 suite of applications (Microsoft Word, Excel and Powerpoint) and IBM SPSS Statistics version 26 (the program in which the statistical analysis was carried out and most of the results obtained).

Chapter 6. Descriptive analysis of the patients enrolled

Introduction. Concerning the descriptive analysis, we established a specific main objective: to perform an assessment of patients with chronic liver disease by analyzing a range of anamnestic, demographic, clinical and biological factors. As a secondary objective, we aimed to identify possible correlations between demographic factors, etiological factors, risk

behaviors, patients' comorbidities and severity of liver disease. At the same time, we compare the differences regarding the variables mentioned above between different stages of chronic liver disease.

Material and method. Demographic, anamnestic, clinical and paraclinical data were collected from all the patients enrolled:

- age;
- gender;
- body mass index/BMI (kg/m^2);
- environment of origin (urban versus rural);
- years of schooling;
- smoking status;
- alcohol consumption (deny/former drinker versus chronic drinker);
- the presence of diabetes mellitus;
- the presence of cardiovascular comorbidities;
- clinical and anamnestic parameters: etiology of liver disease (alcoholic, viral, mixed, associated with non-alcoholic fatty liver or other causes), presence of ascites (clinically/ultrasoun appreciated as absent/minimum amount, medium amount, large amount), history of overt hepatic encephalopathy, ongoing treatment with lactulose and/or rifaximin;
- biological parameters: hemoglobin (g/dL), platelets/μL, serum albumin (g/dL), serum creatinine (mg/dL), total serum bilirubin (mg/dL), serum sodium (mmol/L), AST (U/L), ALT (U/L), INR;
- the presence and type of anemia;
- glomerular filtration rate/GFR (ml/min/1,73 m^2);
- Child-Pugh score;
- MELD-Na score.

Results and discussions. The demographic analysis of the entire study group showed that the patients with chronic liver diseases have an average age of 54.69±9.75 years, the youngest patient included being 27 years old, and the oldest 79 years old. The average body mass index (BMI) in the entire study group was 28.28±3.17 kg/m^2, the average years of

schooling 11.46±2.45, and most of the patients came from the urban environment (138 patients, 77.5%).

In terms of etiology, we observed that the majority of chronic liver diseases were associated with chronic viral hepatitis, either as a single etiology (71 patients, 39.89%) or as a mixed etiology in association with alcohol consumption (22 patients, 12.36%). The second most common etiology was alcohol consumption, either as a single etiology (52 patients, 29.21%) or in association with chronic viral hepatitis (22 patients, 12.3%). Non-alcoholic fatty liver (NAFLD) was encountered in 31 patients (17.42%), and autoimmune etiology was encountered in 2 patients (1.12%). The most common etiology in pre-cirrhotic patients was viral (43.8%), followed by NAFLD (28.15%), alcoholic (22.9%) and mixed etiology (5.2%). The most common etiology in the group of cirrhotic patients was alcoholic (36.6%), followed by viral (35.4%), mixed (20.7%), NAFLD (4.9%) and autoimmune (2.4%).

Also, anamnestically, we assessed habits and comorbidities at risk for chronic liver disease: chronic alcohol consumption, smoking, diabetes, cardiovascular disease. Thus, 66 patients (37.1%) with chronic liver disease confirmed regular alcohol consumption, the remaining patients (112 patients, 62.9%) being either abstinent for more than 3 months, or having categorically denied alcohol consumption or reporting occasional consumption. In addition, 108 patients (60.7%) in the study group were non-smokers or were abstinent for more than 3 months. 61 patients (34.3%) of the enrolled patients had diabetes mellitus (DM), and 58 patients (32.6%) of the total number of subjects had cardiovascular disease.

By comparing between the two groups, we observed that the mean age of patients with cirrhosis is significantly higher than that of patients with chronic liver disease in precirrhotic stages. A significant number of patients with cirrhosis are over 60 years old (48.8%), while the majority of patients with precirrhosis are under 60 years old (84.4%). The BMI of patients with cirrhosis is significantly lower compared to that of patients with chronic hepatopathies in precirrhotic stages. Among patients with cirrhosis the level of education is significantly lower and rural background is more common. Chronic ethanol consumption is significantly more prevalent in patients with liver cirrhosis, while smoking does not differ between the two groups. Diabetes mellitus is more common in cirrhosis compared to precirrhosis, while cardiovascular diseases have similar prevalences. The laboratory parameters followed in the study statistically

differentiated precirrhotic from cirrhotic stages. In patients with cirrhosis, decompensation was observed to occur at significantly higher ages. BMI is significantly lower in decompensated cirrhosis, where chronic ethanol consumption is reported more frequently. The most prevalent etiology for decompensated cirrhosis is alcohol-associated, while for compensated cirrhosis viral etiology predominates. Education level, background, smoking, diabetes mellitus and cardiovascular disease do not differentiate compensated from decompensated cirrhosis.

Chapter 7. The study of neurocognitive dysfunctions

Introduction. In the early stages of minimal hepatic encephalopathy, neurocognitive dysfunctions affect complex activities involving attention, information processing, reaction time and psychoemotional skills. Throughout this chapter, we set as our specific primary objective the assessment of minimal hepatic encephalopathy (MHE) among the patients with chronic liver disease included in the study. As secondary objectives, the study aims to identify factors associated with EHM and its predictors according to the stage of liver disease.

Material and method. We analyzed the entire group of 178 patients enrolled in the study, assessing each patient's cognitive function using the psychometric hepatic encephalopathy score (PHES). This score includes 5 paper-and-pencil tests that assess cognitive/psychomotor processing speed and visual-motor coordination. The following tests were administered:

a) NCT-A and NCT-B=number connection test-A and B.

b) DST=digit symbol test

c) LTT=line-tracing test - divided into two subtests: LTT-t (time) and LTT-e (errors).

d) SDT=serial-dotting test.

Results and discussions. Throughout this study, we analyzed the results of psychometric tests, completed by the entire study group and we performed comparative analyses between the two groups (precirrhosis vs. cirrhosis) and subgroups (compensated cirrhosis vs. decompensated cirrhosis).

Psychometric tests used among patients with chronic liver disease enrolled in the study showed a prevalence of MHE of 34.8%. The independent predictor of MHE among all chronic

liver disease patients was age. By advancing in age with each year, the risk of these patients to present MHE increases by 8%.

In precirrhotic stages, the prevalence of neurocognitive dysfunctions was 21.9% and was associated with a number of factors such as age, years of schooling, background, chronic alcohol consumption, diabetes mellitus, etiology of liver disease and glomerular filtration rate (GFR). Background (rural), chronic alcohol consumption, diabetes mellitus and etiology are independent predictors for MHE in patients with precirrhosis.

In cirrhotic stages, the prevalence of EHM was significantly higher (50%), correlating with the severity of liver cirrhosis. In compensated cirrhosis, the prevalence of neurocognitive dysfunction was 32%, rising to 78.1% in decompensated cirrhosis and was associated with age, chronic alcohol consumption, Child Pugh score, MELD-Na score, history of overt hepatic encephalopathy and RFG. Independent predictors forMHE in patients with cirrhosis are severity of liver cirrhosis and MELD-Na score.

Chapter 8. The study of sleep disorders

Introduction. Sleep pathology has recently become an increasingly intensively studied domain in patients with various chronic diseases. Chronic liver diseases are associated with sleep disturbances, their prevalence varying widely in the literature, from 47% to 81%, due to different methods of assessment, heterogeneity of the population studied, but also due to external factors (e.g., coffee consumption, sleep medication) or associated comorbidities.

The main specific objective of the study of "sleep disorders" was to identify and characterize these disturbances among the patients enrolled with chronic liver diseases, by means of subjective diagnostic methods (sleep questionnaires) and semi-objective methods (actigraphy). The secondary objectives aim, through the comparative analysis, to evaluate the associations between these sleep pathologies and various characteristics of the patients, according to the severity of liver disease. In addition, we aimed to identify predictors of sleep disturbances and the association between sleep parameters and MHE parameters.

Material and method. The sleep of the patients enrolled was assessed by both subjective and semi-objective methods in order to increase diagnostic sensitivity and specificity.

Subjective methods used to assess sleep disorders were represented by the following questionnaires:

- Pittsburgh Sleep Quality Index (PSQI)
- Epworth Sleepiness Scale (ESS)
- STOP-BANG questionnaire (**S**noring, **T**iredness, **O**bserved apnea, blood **P**ressure, **B**ody mass index, **A**ge, **N**eck circumference and **G**ender)
- International Restless Legs Study Group criteria 2014 (IRLSSG), followed by IRLSS severity scale in patients who fulfill IRLSS criteria.

Among the semi-objective methods for evaluating sleep disorders, we used actigraphy. Actigraphy can be considered an alternative to polysomnography, being more cost-effective and easier to use in an outpatient setting. Technically, the actigraph is a wristwatch that has an integrated accelerometer through which the patient's movements are monitored.

For the present research, we had 3 actiwatches, model Actiwatch Philips Respironics; Spectrum Pro produced by Philips Healthcare USA (Figure 2).

Figure 2. Image of the 3 actiwatches used for conduction of the PhD thesis

The data recorded by the actiwatch were analyzed by a computer software that provides a report on sleep parameters (Figure 3). The parameters measured by the actigraph are: bedtime, wake time, time spent in bed, total sleep time, sleep latency, sleep efficacy, wake time after sleep onset (WASO) and number of awakenings per night.

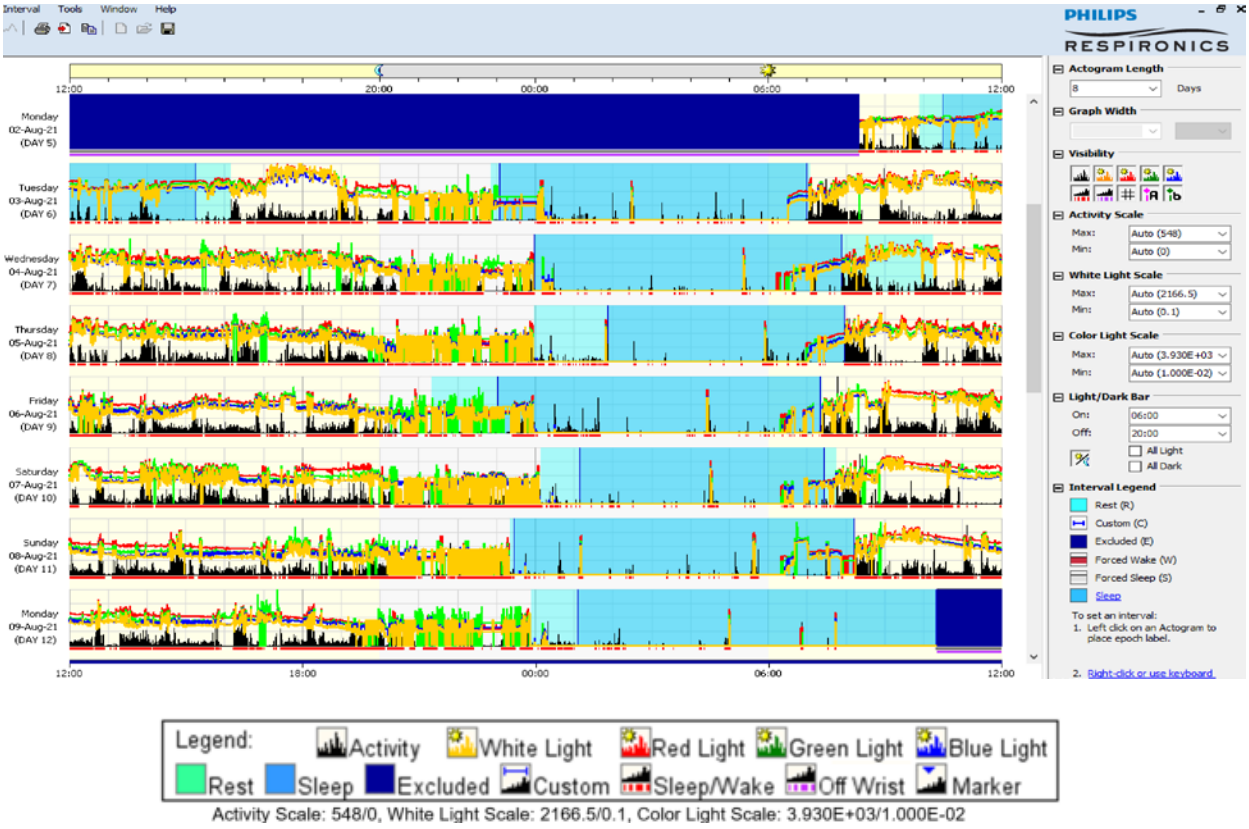

Figure 3. Graphical report with legend

Results and discussions. The sleep disorders encountered among the patients with chronic liver diseases from the present study were insomnia (“poor sleep”), daytime sleepiness, obstructive sleep apnea syndrome (OSA) and restless legs syndrome (RLS).

The prevalence of sleep disturbances during night, translated by unsatisfactory sleep and evaluated by the PSQI questionnaire, was 39.9% among all included patients. The prevalence increased significantly in patients with cirrhosis (50%), rising to 78.1% among decompensated stages. The independent predictors of "poor sleep" among patients with precirrhosis were BMI, diabetes and alcohol etiology, and among patients with cirrhosis, the independent predictors of "poor sleep" were the severity of liver cirrhosis and the presence of MHE.

Daytime sleepiness, assessed by the ESS questionnaire, recorded a prevalence of 29.2%. The frequency increased significantly with the degree of decompensation of the liver disease, reaching 62.5% among decompensated cirrhosis. In the precirrhotic stages, the presence of MHE was an independent predictor for daytime sleepiness. In patients with liver cirrhosis,

independent predictors for daytime sleepiness were chronic alcohol consumption, alcohol and mixed etiology and the presence of MHE.

Increased risk of OSA, as assessed by the STOP-BANG questionnaire, was observed in 26.4% of patients with chronic liver disease, with prevalence rising proportionally with the severity of the disease. The independent predictors of OSA in patients with pre-cirrhosis were found to be age, BMI, the environment of origin and the presence of MHE, and in patients with cirrhosis, the independent predictor for the increased risk of OSA was male gender.

The prevalence of RLS among patients with chronic liver disease was 39.3%, rising with the severity of liver disease to 50% in compensated cirrhosis and 87.5% in decompensated cirrhosis. In patients with precirrhosis, the only independent predictor for RLS was the presence of MHE. In patients with cirrhosis, the independent predictors for RLS were chronic ethanol consumption and low GFR. The presence of anemia was associated with RLS among all patients with chronic liver disease, but there was no association between RLS and the type of anemia. In terms of liver disease etiology, the study results showed no association with the presence of RLS. There was a strong, inversely proportional association between the IRLSS severity score and RFG. The severity of RLS symptoms is directly associated with increased MELD-Na score and liver cirrhosis decompensation.

Strong and moderate correlations were observed between MELD-Na score and PSQI, ESS and STOP-BANG scores. The most important differences were observed in decompensated stages of liver cirrhosis, even in the absence of overt hepatic encephalopathy, which was one of the exclusion criteria.

Actigraphy has shown that patients with cirrhosis fall asleep after significantly longer periods of latency and at later times compared to patients with precirrhosis. Although patients with cirrhosis wake up later and spend more time in bed, the effective sleep period is significantly shorter compared to patients with precirrhosis. In addition, the number of awakenings per night of patients with cirrhosis is significantly higher compared to patients with precirrhosis, and their overall sleep efficacy is lower.

Chronic alcohol use determined patients with chronic liver disease to wake up later, spend significantly more time in bed, and experience fragmented sleep with an increased number of awakenings. Overall sleep efficiency is significantly lower in patients who

chronically consume alcohol compared to patients who are weaned. Smoking, on the other hand, does not statistically differentiate the two groups. Both patients with diabetes and those with cardiovascular disease have lower sleep efficacy, significantly longer sleep latency with multiple waking episodes and longer WASO. In addition to patients with cardiovascular disease, patients with diabetes also spend significantly more time in bed compared to non-diabetics.

The efficacy of sleep assessed by actigraphy correlates strongly and inversely proportionally with the MELD-Na score. Sleep efficiency is significantly lower in patients who have been diagnosed in the past with overt HE and in patients who are receiving treatment with lactulose and/or rifaximin. This last finding might be explained by the fact that most patients undergoing treatment also have advanced stages of cirrhosis, and probably a history of over hepatic encephalopathy.

There were strong associations between sleep efficacy and the presence of IRLSS criteria, moderate associations between sleep efficacy and PSQI and ESS scores, and a modest association with STOP-BANG scores.

Chapter 9. Reassessment of patients at 6 months

Introduction. Throughout the present study, we aimed to reassess patients enrolled at baseline, who were initially assessed by both sleep questionnaires and actiwatch. We investigated the neurocognitive changes that appeared when completing the psychometric tests by comparing the results from the time of enrollment with those from 6 months. In addition, we aimed to analyze the responses to the sleep questionnaires and the parameters recorded by actiwatch after 6 months of disease evolution.

Material and method. After 6 months from enrollment, the patients were contacted by telephone. At the time of the reevaluation, some of the variables monitored at enrollment were retained for statistical comparative analysis, as follows: risk behaviors, BMI, Child Pugh score, MELD-Na score, treatment with lactulose and/or rifaximin. The same psychometric tests and sleep questionnaires completed at the time of enrollment were used. Also, the patients were asked to wear the actiwatch again for 7 consecutive days.

In the 6-month analysis were included 32 patients with precirrhosis and 16 patients with cirrhosis (all with compensated cirrhosis, as those with decompensated cirrhosis either died or refused to be reevaluated in the study).

Results and discussions. Out of the patients with precirrhosis, all remained with precirrhosis after 6 months, but out of the patients with compensated cirrhosis Child A, after 6 months, 3 patients had decompensated Child B cirrhosis and 2 patients had Child C cirrhosis. The MELD-Na score also increased significantly after 6 months, consistent with the decompensation of cirrhosis in the 5 patients.

Minimal hepatic encephalopathy has been associated with increased risk of liver disease progression and could be considered a marker of liver cirrhosis aggressiveness. Of the 31 patients without MHE at enrollment, we observed that after 6 months, 4 patients had MHE, and these were patients with cirrhosis. In the specific analysis of psychometric tests within each group, the most important differences were identified in patients with cirrhosis, whose PHES scores were significantly reduced after 6 months of disease progression. Therefore, we can state that neurocognitive dysfunction progresses in 6 months of evolution of compensated cirrhosis, although apparently most patients remain stable for a long time in this stage of cirrhosis. Out of the 16 patients with compensated cirrhosis who were reevaluated, 14 patients were not undergoing lactulose and/or rifaximin treatment at the time of enrolment. After 6 months, however, of the 14 patients, half were on treatment. As the number of patients reassessed was small, we cannot comment on the effect lactulose and/or rifaximin might have on the progression of MHE in patients with compensated cirrhosis, and future large studies are needed to clarify this issue.

The results obtained when completing the sleep questionnaires after 6 months show a deterioration in sleep quality and an increase in the prevalence of daytime sleepiness, the changes being more significant in patients with cirrhosis. Obstructive sleep apnea syndrome did not change significantly between enrolment and 6 months. Actigraphic reassessment suggests a significant reduction in sleep efficiency, both for precirrhotic patients in whom it remains above the 80% threshold suggestive of effective sleep, but especially for patients with cirrhosis in whom sleep efficiency falls below 80% after 6 months.

Chapter 10. Conclusions and personal contributions

The research objectives, both general and specific to each study, have been achieved and the results lead to the following conclusions:

1. Patients with chronic liver diseases have different demographic, anamnestic, clinical and paraclinical characteristics and some particularities, depending on the stage of liver disease.
2. Neurocognitive dysfunctions occur in the early stages of chronic liver disease and correlates with severity of liver disease, patient comorbidities, risk factors and demographic variables.
3. None of the patients with compensated cirrhosis undergoing treatment with lactulose and/or rifaximin showed neurocognitive dysfunction suggestive of a diagnosis of MHE.
4. In stages of decompensated cirrhosis and in patients with a history of overt hepatic encephalopathy, treatment with lactulose and/or rifaximin did not statistically influence the presence of MHE.
5. As chronic liver disease progresses, the prevalence and severity of sleep-related disorders increase.
6. Sleep characteristics assessed by actigraphy show particularities depending on the stage of liver disease, risk behaviors and associated comorbidities.
7. Sleep efficacy is influenced by MELD-Na score, history of over hepatic encephalopathy and treatment with lactulose and/or rifaximin.
8. Sleep efficacy assessed by actiwatch correlates with subjective parameters assessed by sleep questionnaires.
9. Reassessment of patients at 6 months confirmed that worsening of sleep disturbances and neurocognitive dysfunction occurs with progression of liver disease.

Research limitations. The main limitations of the present research are related to the SARS-COV2 pandemic period, which influenced the relatively small number of patients enrolled, particularly among those reevaluated. Another limitation of the research was the subjective nature of the sleep questionnaires. In addition, only one method was used to detect neurocognitive dysfunction, namely psychometric tests. Although these are considered the gold-standard diagnostic method for MHE, another diagnostic method, such as computerized testing, would certainly have added value to the work. In addition, some parameters were not recorded,

such as: background medication, coffee consumption, reasons for decompensation of liver disease, etc. Last but not least, we should mention the lack of a control group of healthy patients from the general population, without liver disease, which could have helped us as a reference mark for normal results.

Personal contributions and research perspectives. This work contributes to interdisciplinary research in the fields of hepatology, internal medicine and neurosciences thanks to the innovative nature of the methods used, while revealing new research perspectives through the results obtained.

The study of neurocognitive dysfunctions and that of sleep disorders showed that patients with chronic liver diseases present these pathologies from the early stages, even in the absence of overt hepatic encephalopathy and before its setting. The physiopathological mechanisms that determine their appearance remain to be studied in future research.

A peculiarity of the research and at the same time a novelty for Romania is the creation of a website (https://www.phes.ro/), with free access, where doctors will be able to find out after entering the results of the psychometric tests, if their patient presents MHE.

Another innovative contribution of the present research is the use of actigraphy in patients with chronic liver diseases in Romania for the diagnosis of sleep disorders.

Based on the results of the present work, the proposals for the following research consist in expanding the study group of patients with chronic liver diseases and adding a control group of healthy patients, where reevaluations should be carried out at several time intervals. I consider useful and interesting to study of treatments for the prevention and treatment of overt hepatic encephalopathy, in the prevention and treatment of sleep disorders and subclinical neurocognitive dysfunctions in minimal hepatic encephalopathy. Also, analyzing the relationship between sleep disorders, respectively subclinical neurocognitive dysfunctions and the quality of life, the degree of depression and fatigue of patients with chronic liver diseases can represent current and important topics for future research.

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List of published scientific papers

Articles published in journals indexed in Web of Science with impact factor

[1] **Plotogea O-M,** Diaconu CC, Gheorghe G, Stan-Ilie M, Oprita R, Sandru V, Bacalbasa N, Constantinescu C. The Prevalence and Predictors of Restless Legs Syndrome in Patients with Liver Cirrhosis. Healthcare, 10, 822, 2022. https://doi.org/10.3390/healthcare10050822 IF 3.160

[2] **Plotogea O-M,** Diaconu CC, Gheorghe G, Stan-Ilie M, Badea MA, Cijevschi Prelipcean C, Constantinescu G. The Prevalence and Association of Cognitive Impairment with Sleep Disturbances in Patients with Chronic Liver Disease. Brain Sciences, 12, 444, 2022. https://doi.org/10.3390/brainsci12040444 IF 3.333

[3] **Plotogea O-M,** Gheorghe G, Stan-Ilie M, Constantinescu G, Bacalbasa N, Bungau S, Diaconu CC. Assessment of Sleep among Patients with Chronic Liver Disease: Association with Quality of Life. J. Pers. Med., 11, 1387, 2021. https://doi.org/10.3390/jpm11121387 IF 3.508

[4] **Plotogea O-M,** Ilie M, Bungau S, Chiotoroiu AL, Stanescu AMA, Diaconu CC. Comprehensive Overview of Sleep Disorders in Patients with Chronic Liver Disease. Brain Sciences, 11, 142, 2021. https://doi.org/10.3390/brainsci11020142 IF 3.333

[5] **Plotogea O**, Ilie M, Sandru V, Chiotoroiu A, Bratu O, Diaconu C. Cardiovascular and Metabolic Consequences of Liver Transplantation: A Review. Medicina, 55, 489, 2019. https://doi.org/10.3390/medicina55080489 IF 1.205