International Journal of Emerging Trends in Health Sciences

Volume 8, Issue 2, (2024) 36-45

www.ij-hs.eu

Alteration of the gut microbiota in patients with nonalcoholic fatty liver disease and obstructive sleep apnea: a multiplied burden

Doina Georgescu [a*] ,Department of Internal Medicine I, "V Babes" University of Medicine and Pharmacy, Timisoara, Romania.

Daniel Lighezan [b], Department of Internal Medicine I, "V Babes" University of Medicine and Pharmacy, Timisoara, Romania.

Roxana Buzas [c], Department of Internal Medicine I, "V Babes" University of Medicine and Pharmacy, Timisoara, Romania.

Ciprian Ilie Rosca [d], Department of Internal Medicine I, "V Babes" University of Medicine and Pharmacy, Timisoara, Romania.

Oana Elena Ancusa [e], Department of Internal Medicine I, "V Babes" University of Medicine and Pharmacy, Timisoara, Romania.

Ioana Suceava[f], Department of Internal Medicine I, "V Babes" University of Medicine and Pharmacy, Timisoara, Romania

Mihai Ionita [g], Department of Internal Medicine I, "V Babes" University of Medicine and Pharmacy, Timisoara, Romania.

Daniela Reisz [h], Department of Neurosciences, "V Babes" University of Medicine and Pharmacy, Timisoara, Romania.

Suggested Citation:

Georgescu, D., Lighezan, D., Buzas, R., Rosca, C.I., Ancusa, O.E., Suceava, I., Ionita, M.& Reisz, D. (2024). Alteration of the gut microbiota in patients with nonalcoholic fatty liver disease and obstructive sleep apnea: a multiplied burden. *International Journal of Emerging Trends in Health Sciences. 8*(2), 36-45. https://doi.org/10.18844/ijeths.v8i2.9619

Received March 20, 2024; revised from August 8, 2024; accepted from December 2, 2024.
Selection and peer review under the responsibility of Prof. Dr. Nilgun Sarp, Uskudar University, İstanbul.

Abstract

Nonalcoholic fatty liver disease (NAFLD), a globally prevalent condition with an increasing trend, is frequently associated with obstructive sleep apnea (OSA). This study investigated the clinical characteristics and gut microbiota alterations in patients with NAFLD and OSA. Comprehensive evaluations included clinical examinations, body mass index (BMI), blood pressure, laboratory tests, fatty liver index (FLI), liver imaging, cardiovascular assessments, polysomnography, and stool analysis for dysbiosis, with next-generation 16S ribosomal RNA sequencing performed in patients with dysbiosis. Patients with OSA exhibited more advanced liver damage, higher BMI and FLI, and increased prevalence of comorbidities such as diabetes, dyslipidemia, metabolic syndrome, atherosclerotic plaques, ischemic heart disease, diastolic heart failure, and pulmonary hypertension. Significant correlations were observed between the severity of nonalcoholic steatohepatitis (NASH) and dysbiosis, as well as between dysbiosis and FLI. Gut microbiota analysis revealed reduced biodiversity, imbalances in Firmicutes/Bacteroidetes ratios, and altered levels of key microbial species such as Faecalibacterium prausnitzii, Akkermansia muciniphila, and Prevotella spp. Patients with NAFLD and OSA exhibited more severe liver damage, increased metabolic and cardiovascular risks, and notable gut microbiota alterations. These findings suggest that gut microbiota assessment may offer valuable insights for tailored therapeutic interventions to improve patient outcomes.

Keywords: Nonalcoholic fatty liver disease; obstructive sleep apnea; gut microbiota alterations.

* ADDRESS FOR CORRESPONDENCE: Doina Georgescu,Department of Internal Medicine I, "V Babes" University of Medicine and Pharmacy, Timisoara, Romania.
E-mail address: doina.georgescu@umft.ro

Georgescu, D., Lighezan, D., Buzas, R., Rosca, C.I., Ancusa, O.E., Suceava, I., Ionita, M.& Reisz, D. (2024). Alteration of the gut microbiota in patients with nonalcoholic fatty liver disease and obstructive sleep apnea: a multiplied burden. *International Journal of Emerging Trends in Health Sciences. 8*(2), 36-45. https://doi.org/10.18844/ijeths.v8i2.9619

1. INTRODUCTION

Nonalcoholic fatty liver disease (NAFLD) is a condition with an upward trend and worldwide distribution, frequently associated with metabolic syndrome, obesity-induced inflammation, and insulin resistance. NAFLD is characterized by the accumulation of triglycerides in more than 5% of liver cells, not associated with alcohol abuse or other etiologies of liver damage. NAFLD may exhibit various clinical and pathological features from simple steatosis, to more severe liver conditions such as nonalcoholic steatohepatitis (NASH) with hepatocyte necrosis and inflammation, or NASH-related cirrhosis in patients with end-stage chronic liver disease where severe fibrosis and architectural reshape are present (Younossi, Koenig, Abdelatif, Fazel, Henry & Wymer, 2016; Chalasani, Younossi, Lavine, Charlton, Cusi, Rinella, Harrison, Brunt & Sanyal, 2018).

NASH is seen as a multifactorial condition, with many factors either genetic or environmental being involved in the disease progression. The shift from steatosis to inflammation and hepatocyte necrosis has been actively studied in recent years. Among many possible triggers associated with the progression to more severe liver damage, gut microbiota dysbiosis is often reported. A lot of recent studies emphasized that NASH was linked to alterations in the intestinal microbiota. As a consequence, the particularities of the intestinal microbiome displayed by the patients with NASH were intensively analyzed (Boursier, Mueller, Barret, Machado, Fizanne, Araujo-Perez, Guy, Seed, Rawls, David, Hunault, Oberti, Calès & Diehl, 2016; Brandl, & Schnabl, 2017; Kobayashi, Iwaki, Nakajima, Nogami, & Yoneda, 2022; Fang, Yu, Li, Yao, Fang, Yoon, and Yu, 2022).

Multiple studies have reported the association between NASH and obstructive sleep apnea (OSA), especially in patients where various bariatric surgical procedures were performed (Asfari, Niyazi, Lopez, Dasarathy & McCullough, 2017; Hany, Abouelnasr, Abdelkhalek, Ibrahim, Aboelsoud, Hozien & Torensma, 2023). Furthermore, recent studies have highlighted that OSA and chronic intermittent hypoxia might be considered risk factors in NAFLD (Tang, Lv, Zhang, Liu & Mao, 2023). The present study aimed to highlight the clinical and gut microbiota particularities in patients with NAFLD and OSA.

2. METHOD AND MATERIALS

2.1. Participants

100 patients, 58 females, and 42 males, age range 29-74 years, with NAFLD were consecutively included in this cross-sectional study, depending on the presence or absence of OSA. A comprehensive clinical evaluation was conducted, including neck and waist measurements, body mass index (BMI), blood pressure assessment, laboratory investigations, fatty liver index (FLI) calculation, liver imaging with abdominal duplex ultrasonography and elastography, Carotid Eco Doppler, ECG, echocardiography, thoracic radiography, polysomnography, and stool microbiology for dysbiosis severity assessment. Next-generation 16S ribosomal RNA sequencing was performed in patients with dysbiosis.

Inclusion criteria: patients diagnosed with NAFLD, with no fibrosis (F0) or mild to moderate fibrosis, ranging from F1 to F3 scoring system.

Exclusions criteria: evidence of heavy alcohol drinking more than 40gr/day in men and 30gr/day in women, over the past 10 years, history of exposure to industrial toxins or to hepatotoxic drugs; various inherited or acquired liver conditions such as Wilson disease, hemochromatosis, alpha1 antitrypsin deficiency, primary biliary cirrhosis, autoimmune hepatitis; hepatic viral infections: HBV, HDV or HCV; organ failure (heart, lung, kidney or liver), recent trauma and surgery, cancer, burning, shock, stroke, bacterial infectious diseases and sepsis, pancreatic diseases, thyroid diseases, long-standing parenteral nutrition, bowel inflammatory disease

and other entities resulting in malnutrition syndromes, recent covid infection, as well as recent treatment with antibiotics or probiotics.

2.2. Procedure

Imaging evaluation of NAFLD was performed for each participant using high-resolution real-time duplex ultrasonography. The severity of liver steatosis was semiquantitatively assessed as follows: mild (1 point), moderate (2 points), and severe (3 points) (Tamaki, Ajmera, & Loomba, 2022). The evaluation of fibrosis was done by using point shear wave elastography. Liver fibrosis was scored as follows F0 no fibrosis, F1F2 mild fibrosis, F3 moderate fibrosis, and E4 severe fibrosis (Loomba, 2018). Abdominal helical CT was also performed in all study participants with the highlight of the decrease of the liver density secondary to lipid accumulation (Li, Dhyani, Grajo, Sirlin & Samir, 2018).

Noninvasive assessment of NAFLD and NASH: Fibromax(BioPredictive®) was used to assess NashTest® and FibroTest® in the study population Fatty liver index (FLI) was calculated using online MedCalc's calculators, based on the waist circumference (cm), BMI (kg/m2), GGT (U/l) and triglycerides (mg/dl) (Piazzolla, & Mangia, 2020; Paul, 2020).

Body mass index BMI was calculated using online MedCalc calculators, based on height(cm) and weight(kg).

Blood pressure assessment Measurements of the blood pressure were performed in the morning, with patients at rest, in a sitting position.

Assessments of diabetes mellitus (DM) or impaired glucose tolerance (IGT) were made according to the criteria of the American Diabetes Association (2014).

Assessment of dyslipidemia was made according to the International Guidelines for the Management of Dyslipidemia (Aygun & Tokgozoglu, 2022).

Assessment of chronic kidney disease (CKD) was made according to the Kidney Disease Improving Global Outcomes (KDIGO), CKD Work Group. KDIGO (2024).

Assessment of OSA was performed according to modified from International Classification of Sleep Disorders – Third edition (ICSD-3) (A and B) or C (Goyal & Johnson, 2017).

Assessment of heart failure was done in accordance with the recommendations of the European Society of Cardiology (ESC) on the diagnosis of heart failure issued in 2021 (McDonagh, et. al., 2021).

Gut microbiota dysbiosis assessment was made after the initial identification of the microorganisms, using matrix-assisted laser desorption ionization-time of flight –mass spectrometry (MALDI-TOF-MS) method and expressed as colony formatting units (CFU)/gram stool (Tran, Alby, Kerr, Jones & Gilligan, 2015). The gut dysbiosis (DB) was considered when a quantitative or qualitative imbalance of the intestinal flora developed. The severity of the gut microbiota DB was scored as follows: 0=absent, 1=mild, 2=medium, 3=severe. If intestinal DB was diagnosed, stools were further analyzed by 16S r RNA next-generation sequencing method (Ji & Nielsen, 2015).

2.3. Statistical analysis

Statistical analysis was performed using Graph Pad Prism ver.10.4.0 (621) software (Graph Pad Software, Inc., La Jolla, CA, USA). Quantitative variables were expressed as mean values (MV) ± standard deviation (SD).

Chi-squared test was used for categorical variables, expressed as percentages. For continuous data, the unpaired t-test was calculated and $p \leqslant 0.05$ was considered statistically significant, with confidence interval CI=95%. The frequency distribution of the data was represented by histograms. The nonparametric Pearson's correlation test was performed and the relationship between variables was represented by graphs.

2.4. Statistical analysis

This study was conducted in accordance with the Declaration of Helsinki, and approved by the Ethical Committee of the Scientific Research of the University of Medicine and Pharmacy from Timisoara, Romania, number 15 a/10.05.2021 extended 22.08.2024.

3. RESULTS

This is a cross-sectional, observational study involving 100 research participants with NAFLD, evenly divided into two groups depending on the presence or absence of OSA. Table 1 presents the demographic data of the patients included in this study.

Table 1.

Baseline demographic data in research participants

Demographic data	OSA (+)	OSA (-)	p
Age	57.7±5.85	53.45±5.05	0.073*
Gender F/M	27/23	32/28	0.3118
Urban residency	74%	62%	0.2006

Legend: OSA= obstructive sleep apnea, F/M=females/males, *= statistically significant

As observed in Table 1, significant differences were noted between the two groups only related to the age of patients, in favor of the OSA+ group. Table 2 illustrates various aspects of the biological and clinical baseline data of the research participants.

Table 2.

Biological and clinical baseline data of the study patients

Variables	OSA(+)	OSA(-)	p
ALT(U/L)	38.51±17.1	25.48±13.5	0.0001*
Creatinin(mg/dl)	0.9±0.23	0.88±0.12	0.5869
CRP(g/dl)	1.072±0.86	0.62±0.53	<0.0001*
Smoking history	52%	40%	0.2310
Sedentary lifestyle	60%	52%	0.4227
FLI(uU)	63.48±7.23	52.23±0.65	<0.0001*
BMI>30kg/m^2	33.03±2.51	29.5±3.21	<0.0001*
Fibrosis F0-F2	66%	78%	0.1836
Fibrosis F3	44%	22%	0.0199*
NASH score	0.28±0.16	0.19±0.08	p=0.0001
G-I associated conditions	56%	48%	0.4257
GSD	36%	32%	0.6744
T2DM/IGT	30%	12%	0.0466*
Dyslipidemia	76%	56%	0.0357*
Metabolic syndrome	66%	40%	0.0001*

C-V conditions	56%	44%	0.2325
CKD	30.0%	22%	0.3642
Gut DB severity	2.31±0.58	1.83±0.51	p<0.0001*

Table 2: Legend OSA= obstructive sleep apnea, ALT=alanine-aminotransferase, CRP=C reactive protein, FLI=fatty liver index, BMI=body mass index, NASH=nonalcoholic steatohepatitis, G-I= gastro-intestinal, GSD= gallstone disease, T2DM/IGT= type2 diabetes mellitus/impaired glucose tolerance, C-V= cardio-vascular, CKD=chronic kidney disease, DB =dysbiosis, *= statistically significant

As seen in Table 2, significant differences in favor of patients with OSA were noted related to age (57.7±5.85 vs. 53.45±5.05, p=0.073), NASH score (0.28±0.16vs. 0.19±0.08, p=0.0001), FLI (63.48±7.23 vs. 52.23±0.65, p<0.0001), liver fibrosis F3 (44%vs.22%, p=0.199), BMI (33.03±2.51vs. 29.5±3.21,p<0.0001), metabolic syndrome (66%vs.40%,p=0.0001), gut dysbiosis (2.31±0.58vs1.83±0.51,p<0.0001) and various comorbidities: diabetes mellitus (30% vs.12%, p=0.0279), dyslipidemia (76%vs.56%,p=0.0357).

However, no significant differences were observed related to chronic kidney disease (40%vs.22%p=0.0528), cardiovascular conditions (56%vs.44%, p=0.2325), gallstone and gastrointestinal diseases (36% vs.32%, p=0.9744 and 56% vs.48%, p=0.4257), low-grade fibrosis F1F2 (66%vs.78%, p=0.1836), as well as lifestyle particularities such as smoking and sedentarism (52%vs.40%, p=0.2310 and 60%vs.52%, p=0.4227).

Table 3 presents the associated cardio-vascular issues analyzed in NAFLD research participants.

Table 3.

Cardio-vascular features in the research participants

Variables	OSA(+)	OSA(-)	p
HT	58%	30%	0.0732
Carotid atherosclerotic plaques	60%	40%	0.0466*
LVEF	52.05± 4.85	53.45± 5.05	0.1606
Diastolic heart insufficiency	54%	34%	0.045*
Pulmonary hypertension	36%	6%	0.0002*
Ischemic heart disease	60%	34%	0.0096*

Table 3 Legend OSA= obstructive sleep apnea, HT= arterial hypertension, KVED =left ventricular ejection fraction,*= statistically significant

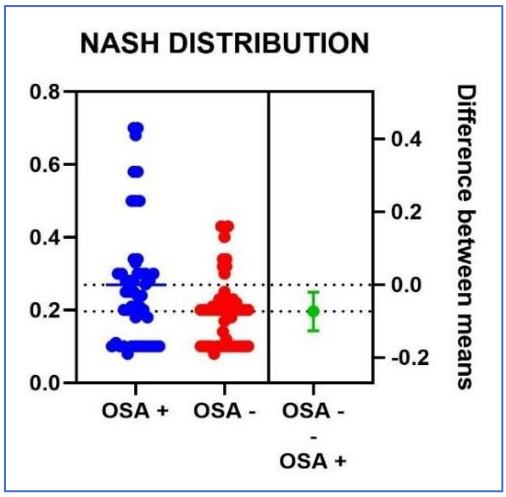
As illustrated in Table 3, significant differences between the two groups of research participants in relation to cardio-vascular comorbidities were observed in favor of the OSA+ group in connection with the atherosclerotic plaques (60%vs.40%, p=0.0466), diastolic heart insufficiency (54%vs.34%, p=0.045), pulmonary hypertension (36%vs.6%, p=0.0002) and ischemic heart disease (60% vs. 34%, p=0.0086).

However, no significant differences were noted related to arterial hypertension (58%vs.40%, p=0.0732) and LVEF (52.05± 4.85vs. 53.45± 5.05 p=0.1606).

The NASH distribution of the two groups, analyzed by the present study is illustrated in figure 1.

Figure 1.

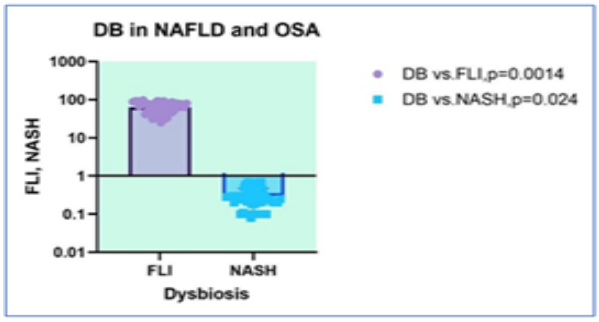
NASH distribution in patients OSA + vs. OSA-



As observed in Figure 1, close correlations between NASH scores and OSA severity were noted. Figure 2 displays the correlations between dysbiosis range, FLI, and NASH severity in patients with OSA (+).

Figure 2.

Correlations between DB severity, FLI, and NASH score



As illustrated in Figure 2, close correlations between NASH and dysbiosis severity (r= 0.3187), as well as between DB and FLI (r=0.4396) were observed in the OSA+ group.

Table 4. depicts the distribution of the gut microbiota DB in research participants diagnosed with NAFLD and associated OSA.

Table 4.

Particularities of gut microbiota DB in OSA (+) research participants

DB Variables	OSA+
Overall DB score	2.31±0.58
Imbalance of F/B	84%
Increased F/B	8%
Decreased F/B	76%
Firmicutes/Bacteroidetes (F/B)	2.64±0.76
Shannon-Wiener H index	2.72±0.42
Decreased Shannon- Wiener H index	76%

Enterotype dominant Prevotella spp.	84%
Enterotype	16%
Increased LPS (+) bacteria spp.	76%
Decreased Akkermansia muciniphila spp.	60%
Decreased Faecalibacterium prausnitzii	66%

As observed in Table 4, a decrease in stool biodiversity, abundance of enterotypes 1 and 2, alterations of Firmicutes/Bacteroidetes ratio, and modifications of functional microbiota, such as Faecalibacterium prausnitzii, Akkermansia muciniphila, and LPS (+) bacteria spp. as well as Prevotella spp., were frequently noted in patients with NAFLD and OSA.

4. DISCUSSION

It is by now already proven that gut microbiota homeostasis plays a key role in maintaining the health of the humans and of the animals, by modulating various steps of the host metabolism. Furthermore, the imbalance of the gut microbiome may trigger numerous pathways that will affect immunity or favor inflammation.

Over the past decade, a lot of studies have associated the gut microbiota DB to several functional conditions such as irritable bowel syndrome, or pathologies beyond the gastrointestinal tract like migraine and other chronic pain conditions (Georgescu, et. al., 2019; Lin, Wang, Zhang, Yuan, Zhang & Chen, 2020; Garvey, 2023; Kashyap & Wang, 2024; Kashyap & Wang, 2024; Goudman, 2024).

A growing body of recent literature linked the gut microbiota DB to a multitude of organic diseases not only metabolic ones, including obesity and insulin resistance, diabetes, and atherosclerosis, but also to liver and kidney diseases, as well as to various neurogenerative conditions (Feng, Wang, Dong, Jiang, Zhang, Raza & Lei, 2021; Georgescu, Ancusa, Azoulay, Lascu, Ionita, Calamar-Popovici, Ionita, Rosca, Breazǎ, Reisz, & Lighezan, 2023; Denman, Park & Jo, 2023; Intili, Paladino, Rappa, Alberti, Plicato, Calabrò, Fucarino, Cappello, Bucchieri, Tomasello, Carini, & Pitruzzella, 2023).

The metabolic syndrome and NAFLD are often reported to be associated with alterations of the gut microbiota DB. Note that NAFLD and OSA share numerous pathogenic conditions including the gut microbiota imbalance, which is ubiquitous observed in both situations (Durgan, Ganesh, Cope, Ajami, Phillips, Petrosino, Hollister & Bryan, 2016; Mashaqi & Gozal, 2019; Bikov, Szabo, Piroska, Kunos, Szily, Ligeti, Makra, Szabo, Tarnoki & Tarnoki, 2022; Munir, Sert Kuniyoshi, Singh & Covassin, 2023; Li, Xu, Shao, Gao, Wang, Dong, Wang, Lu, Li, Tan, Jiang, Xie, Cai, Feng & Li, 2023). The observations of the present study highlighted that the patients with NAFLD and OSA displayed clinical particularities characterized by older age and more severe NASH and FLI scores. The incidence of metabolic conditions was significantly higher compared to the group who did not exhibit OSA. Also, cardio-vascular comorbidities were more frequently encountered in NAFLD patients having OSA.

Markers of inflammation such as CRP were significantly elevated in patients with NAFLD and OSA, being correlated to the severity of the DB range. Dysbiotic patients from the study group exhibited a higher amount from the LPS (+) bacteria spp, known to trigger various proinflammatory cytokines and interleukins. Alterations of the gut microbiota biodiversity, a decrease of the mucosa protective and mucin degrading bacteria, with diminution of Akkermansia muciniphila spp. and Faecalibacterium prausnitzii spp. were also observed in OSA

+ patients. Abundance of the enterotype Prevotella spp. dominant, noted in the OSA+ group might be also associated with lifestyle particularities with sedentary life and low fiber diet, observed in these patients.

The observations related to microbiota changes in patients having not only NAFLD but also OSA may provide valuable keys not only for understanding what is the real dimension of the personal and social burden but also for drawing supporting strategies to improve their outcomes.

5. CONCLUSIONS

Patients with NAFLD and OSA displayed older age, more severe liver damage, increased average BMI and FLI, as well as numerous cardiovascular and metabolic issues. Alterations of the gut microbiota biodiversity, decreased mucosa protection, and mucin-degrading bacteria represent an additional burden, multiplying the risk of bad outcomes. The assessment of microbiota changes may provide valuable keys for customized interventions to improve the outcomes.

REFERENCE

American Diabetes Association (2014). Diagnosis and Classification of Diabetes Mellitus. Diabetes Care, 37, S81-S90. https://doi.org/10.2337/dc14-S081

Asfari, M. M., Niyazi, F., Lopez, R., Dasarathy, S., & McCullough, A. J. (2017). The association of nonalcoholic steatohepatitis and obstructive sleep apnea. *European journal of gastroenterology & hepatology*, *29*(12), 1380–1384. https://doi.org/10.1097/MEG.0000000000000973

Aygun, S., & Tokgozoglu, L. (2022). Comparison of Current International Guidelines for the Management of Dyslipidemia. *Journal of clinical medicine*, *11*(23), 7249. https://doi.org/10.3390/jcm11237249

Bikov, A., Szabo, H., Piroska, M., Kunos, L., Szily, M., Ligeti, B., Makra, N., Szabo, D., Tarnoki, D. L., & Tarnoki, A. D. (2022). Gut Microbiome in Patients with Obstructive Sleep Apnoea. *Applied Sciences*, *12*(4), 2007. https://doi.org/10.3390/app12042007

Boursier, J., Mueller, O., Barret, M., Machado, M., Fizanne, L., Araujo-Perez, F., Guy, C. D., Seed, P. C., Rawls, J. F., David, L. A., Hunault, G., Oberti, F., Calès, P., & Diehl, A. M. (2016). The severity of nonalcoholic fatty liver disease is associated with gut dysbiosis and shift in the metabolic function of the gut microbiota. *Hepatology (Baltimore, Md.)*, *63*(3), 764–775. https://doi.org/10.1002/hep.28356

Brandl, K., & Schnabl, B. (2017). Intestinal microbiota and nonalcoholic steatohepatitis. *Current opinion in gastroenterology*, *33*(3), 128–133. https://doi.org/10.1097/MOG.0000000000000349

Chalasani, N., Younossi, Z., Lavine, J. E., Charlton, M., Cusi, K., Rinella, M., Harrison, S. A., Brunt, E. M., & Sanyal, A. J. (2018). The diagnosis and management of nonalcoholic fatty liver disease: Practice guidance from the American Association for the Study of Liver Diseases. *Hepatology (Baltimore, Md.)*, *67*(1), 328–357. https://doi.org/10.1002/hep.29367

Denman, C. R., Park, S. M., & Jo, J. (2023). Gut-brain axis: gut dysbiosis and psychiatric disorders in Alzheimer's and Parkinson's disease. *Frontiers in neuroscience*, *17*, 1268419. https://doi.org/10.3389/fnins.2023.1268419

Durgan, D. J., Ganesh, B. P., Cope, J. L., Ajami, N. J., Phillips, S. C., Petrosino, J. F., Hollister, E. B., & Bryan, R. M., Jr (2016). Role of the Gut Microbiome in Obstructive Sleep Apnea-Induced Hypertension. *Hypertension (Dallas, Tex.:1979)*, *67*(2),469–474. https://doi.org/10.1161/HYPERTENSIONAHA.115.06672

Georgescu, D., Lighezan, D., Buzas, R., Rosca, C.I., Ancusa, O.E., Suceava, I., Ionita, M.& Reisz, D. (2024). Alteration of the gut microbiota in patients with nonalcoholic fatty liver disease and obstructive sleep apnea: a multiplied burden. *International Journal of Emerging Trends in Health Sciences*. *8*(2), 36-45. https://doi.org/10.18844/ijeths.v8i2.9619

Fang J, Yu C-H, Li X-J, Yao J-M, Fang Z-Y, Yoon S-H and Yu W-Y (2022). Gut dysbiosis in nonalcoholic fatty liver disease: pathogenesis, diagnosis, and therapeutic implications. *Front. Cell. Infect. Microbiol.* 12:997018. doi: 10.3389/fcimb.2022.997018

Feng, Z., Wang, T., Dong, S., Jiang, H., Zhang, J., Raza, H. K., & Lei, G. (2021). Association between gut dysbiosis and chronic kidney disease: a narrative review of the literature. *The Journal of international medical research*, *49*(10), 3000605211053276. https://doi.org/10.1177/03000605211053276

Garvey, M. (2023). The Association between Dysbiosis and Neurological Conditions Often Manifesting with Chronic Pain. *Biomedicines*, *11*(3), 748. https://doi.org/10.3390/biomedicines11030748

Georgescu, D., Ancusa, O. E., Azoulay, D., Lascu, A., Ionita, I., Calamar-Popovici, D., Ionita, M., Rosca, C. I., Brează, G. M., Reisz, D., & Lighezan, D. (2023). Portal Vein Thrombosis in Patients with Liver Cirrhosis: What Went Wrong?. *International journal of general medicine*, *16*, 3889–3906. https://doi.org/10.2147/IJGM.S413438

Georgescu, D., Iurciuc, M.S., Petre, I., Georgescu, L.A., Szasz, F., Ionita, I.,... Lighezan, D. (2019). Chronic Pelvic Pain and Irritable Bowel Syndrome: Is Subclinical Inflammation Bridging the Gap?. Revista de Chimie, 70(10), 3634-3637. https://doi.org/10.37358/RC.19.10.7611

Goudman, L., Demuyser, T., Pilitsis, J. G., Billot, M., Roulaud, M., Rigoard, P., & Moens, M. (2024). Gut dysbiosis in patients with chronic pain: a systematic review and meta-analysis. *Frontiers in immunology*, *15*, 1342833. https://doi.org/10.3389/fimmu.2024.1342833

Goyal, M., & Johnson, J. (2017). Obstructive Sleep Apnea Diagnosis and Management. *Missouri medicine*, *114*(2), 120–124..

Hany, M., Abouelnasr, A. A., Abdelkhalek, M. H., Ibrahim, M., Aboelsoud, M. R., Hozien, A. I., & Torensma, B. (2023). Effects of obstructive sleep apnea on non-alcoholic fatty liver disease in patients with obesity: a systematic review. *International journal of obesity (2005)*, *47*(12), 1200–1213. https://doi.org/10.1038/s41366-023-01378-2

Intili, G., Paladino, L., Rappa, F., Alberti, G., Plicato, A., Calabrò, F., Fucarino, A., Cappello, F., Bucchieri, F., Tomasello, G., Carini, F., & Pitruzzella, A. (2023). From Dysbiosis to Neurodegenerative Diseases through Different Communication Pathways: An Overview. *Biology*, *12*(2), 195. https://doi.org/10.3390/biology12020195

Ji, B., & Nielsen, J. (2015). From next-generation sequencing to systematic modeling of the gut microbiome. *Frontiers in genetics*, *6*, 219. https://doi.org/10.3389/fgene.2015.00219

Kashyap, Y., & Wang, Z. J. (2024). Gut microbiota dysbiosis alters chronic pain behaviors in a humanized transgenic mouse model of sickle cell disease. *Pain*, *165*(2), 423–439. https://doi.org/10.1097/j.pain.0000000000003034

Kidney Disease: Improving Global Outcomes (KDIGO) CKD Work Group (2024). KDIGO 2024 Clinical Practice Guideline for the Evaluation and Management of Chronic Kidney Disease. *Kidney international*, *105*(4S), S117–S314. https://doi.org/10.1016/j.kint.2023.10.018

Kobayashi, T., Iwaki, M., Nakajima, A., Nogami, A., & Yoneda, M. (2022). Current Research on the Pathogenesis of NAFLD/NASH and the Gut-Liver Axis: Gut Microbiota, Dysbiosis, and Leaky-Gut Syndrome. *International journal of molecular sciences*, *23*(19), 11689. https://doi.org/10.3390/ijms231911689

Li, Q., Dhyani, M., Grajo, J. R., Sirlin, C., & Samir, A. E. (2018). Current status of imaging in nonalcoholic fatty liver disease. *World journal of hepatology*, *10*(8), 530–542. https://doi.org/10.4254/wjh.v10.i8.530

Georgescu, D., Lighezan, D., Buzas, R., Rosca, C.I., Ancusa, O.E., Suceava, I., Ionita, M.& Reisz, D. (2024). Alteration of the gut microbiota in patients with nonalcoholic fatty liver disease and obstructive sleep apnea: a multiplied burden. *International Journal of Emerging Trends in Health Sciences*. *8*(2), 36-45. https://doi.org/10.18844/ijeths.v8i2.9619

Li, Q., Xu, T., Shao, C., Gao, W., Wang, M., Dong, Y., Wang, X., Lu, F., Li, D., Tan, H., Jiang, Y., Xie, Q., Cai, F., Feng, L., & Li, T. (2023). Obstructive sleep apnea is related to alterations in fecal microbiome and impaired intestinal barrier function. *Scientific reports*, *13*(1), 778. https://doi.org/10.1038/s41598-023-27784-0

Lin, B., Wang, Y., Zhang, P., Yuan, Y., Zhang, Y., & Chen, G. (2020). Gut microbiota regulates neuropathic pain: potential mechanisms and therapeutic strategy. *The journal of headache and pain*, *21*(1), 103. https://doi.org/10.1186/s10194-020-01170-x

Loomba, R. (2018). Role of imaging-based biomarkers in NAFLD: Recent advances in clinical application and future research directions. *Journal of hepatology*, *68*(2), 296–304. https://doi.org/10.1016/j.jhep.2017.11.028

Mashaqi, S., & Gozal, D. (2019). Obstructive Sleep Apnea and Systemic Hypertension: Gut Dysbiosis as the Mediator?. *Journal of clinical sleep medicine : JCSM : official publication of the American Academy of Sleep Medicine*, *15*(10), 1517–1527. https://doi.org/10.5664/jcsm.7990

McDonagh, T. A., Metra, M., Adamo, M., Gardner, R. S., Baumbach, A., Böhm, M., Burri, H., Butler, J., Čelutkienė, J., Chioncel, O., Cleland, J. G. F., Coats, A. J. S., Crespo-Leiro, M. G., Farmakis, D., Gilard, M., Heymans, S., Hoes, A. W., Jaarsma, T., Jankowska, E. A., Lainscak, M., … ESC Scientific Document Group (2021). 2021 ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure. *European heart journal*, *42*(36), 3599–3726. https://doi.org/10.1093/eurheartj/ehab368

Munir, S. S., Sert Kuniyoshi, F. H., Singh, P., & Covassin, N. (2023). Is the Gut Microbiome Implicated in the Excess Risk of Hypertension Associated with Obstructive Sleep Apnea? A Contemporary Review. *Antioxidants (Basel, Switzerland)*, *12*(4), 866. https://doi.org/10.3390/antiox12040866

Paul, J. (2020). Recent advances in non-invasive diagnosis and medical management of non-alcoholic fatty liver disease in adult. *Egypt Liver Journal*, 10(37). https://doi.org/10.1186/s43066-020-00043-x

Piazzolla, V. A., & Mangia, A. (2020). Noninvasive Diagnosis of NAFLD and NASH. *Cells*, *9*(4), 1005. https://doi.org/10.3390/cells9041005

Tamaki, N., Ajmera, V., & Loomba, R. (2022). Non-invasive methods for imaging hepatic steatosis and their clinical importance in NAFLD. *Nature reviews. Endocrinology*, *18*(1), 55–66. https://doi.org/10.1038/s41574-021-00584-0

Tang, H., Lv, F., Zhang, P., Liu, J., & Mao, J. (2023). The impact of obstructive sleep apnea on nonalcoholic fatty liver disease. *Frontiers in Endocrinology*, *14*, 1254459. https://doi.org/10.3389/fendo.2023.1254459

Tran, A., Alby, K., Kerr, A., Jones, M., & Gilligan, P. H. (2015). Cost Savings Realized by Implementation of Routine Microbiological Identification by Matrix-Assisted Laser Desorption Ionization-Time of Flight Mass Spectrometry. *Journal of clinical microbiology*, *53*(8), 2473–2479. https://doi.org/10.1128/JCM.00833-15

Younossi, Z. M., Koenig, A. B., Abdelatif, D., Fazel, Y., Henry, L., & Wymer, M. (2016). Global epidemiology of nonalcoholic fatty liver disease-Meta-analytic assessment of prevalence, incidence, and outcomes. *Hepatology (Baltimore, Md.)*, *64*(1), 73–84. https://doi.org/10.1002/hep.28431