Spectrum Journal of Innovation, Reforms and Development
Volume 19, September, 2023 | ISSN (E): 2751-1731
Website: www.sjird.journalspark.org

SIGNIFICANCE OF BIOMARKERS IN THE DIAGNOSIS OF NON-ALCOHOLIC FATTY LIVER DISEASE

Parpibaeva D. A.
Musaeva M. A.
Tashkent Medical Academy

Abstract
Non-alcoholic fatty liver disease (NAFLD) is considered one of the leading causes of chronic liver disease worldwide. A liver biopsy is required to confirm the diagnosis of the disease, but due to the invasiveness of this method, this cost is not suitable for mass screening. Existing laboratory tests do not adequately meet the requirements for early detection of the pathological process during the initial examination of patients with suspected NAFLD.
At the same time, it is very important to identify patients in the early stages of the development of NAFLD. In recent years, the attention of researchers has been focused on expanding knowledge about the mechanism of NAFLD development and new diagnostic methods. According to the results of the research, the development and progression of NAFLD is regulated by epigenetic factors, in particular, by the family of microribonuclein acids (miRNA, miR), which, in turn, have high diagnostic and prognostic value.
This article reviews the PubMed databases, discusses the potential role of microRNAs in hepatic lipid metabolism, and their importance in the pathogenesis of nonalcoholic fatty liver disease.
The possibility of using microRNAs (miRNA-16, miRNA-21, miRNA-34a, miRNA-103, miRNA-122, miRNA-145, miRNA-192 and other types) as promising biomarkers for minimally invasive diagnosis of NAFLD, assessment of the level and stage of activity of steatosis and fibrosis, and as prognostic markers of the diseases are considered.

Keywords: Non-alcoholic fatty liver disease, liver steatosis, non-alcoholic steatohepatitis, biomarker, microRNA, microelement Zn.

Introduction

Nonalcoholic fatty liver disease encompasses a spectrum of clinical and morphologic conditions ranging from asymptomatic steatosis to nonalcoholic steatohepatitis and liver fibrosis, cirrhosis, and, in some cases, hepatocellular carcinoma. [1]. Hepatic steatosis occurs as a result of excessive absorption of free fatty acids by hepatocytes, for example, in cases of metabolic syndrome and insulin resistance. In addition, biochemical disturbances, oxidative stress, increased synthesis of inflammatory mediators, and apoptosis can lead to liver inflammation and fibrosis [2]. At present, liver biopsy is the "gold standard" in evaluating the histological status of the liver in patients with NAFLD. However, due to the fact that these examination methods have a number of disadvantages [3], there is a growing

interest in finding alternative non-invasive and minimally invasive methods [4] that are effective for the assessment of NAFLD, inflammation and fibrosis level [5].
Clinical and laboratory criteria available in practice do not sufficiently meet the requirements for timely diagnosis of the disease, therefore, new promising research is being conducted that allows to identify the pathological process at an early stage.
This article focuses on the diagnostic value of the family of microribonuclein acids in the diagnosis of patients with NAFLD.

Clinical signs and diagnosis

NAFLD is usually asymptomatic and is often diagnosed incidentally when examining patients for other reasons. Often, NAFLD is accidentally detected when patients turn to the doctor with arterial hypertension, ischemic heart disease, peripheral vascular disease, obesity, type 2 diabetes mellitus and other diseases. Cytolysis syndrome and/or hepatomegaly may be suspected in such patients.
Clinically, symptoms typical of metabolic syndrome may come to the fore: visceral obesity, signs of glucose metabolism disorders, dyslipidemia, and arterial hypertension. Some patients complain of non-specific, i.e. rapid fatigue, stabbing pains or discomfort under the right rib not associated with eating. When NAFLD leads to the development of cirrhosis of the liver, symptoms characteristic of liver failure or portal hypertension appear: abdominal enlargement, edema, hemorrhagic syndrome, encephalopathy and etc. [6, 7].
In case of accidental detection of steatosis, family and personal history should be studied in detail, with the exception of secondary steatosis. Examination of the patient should include a complete assessment of all components of the metabolic syndrome. If a patient has obesity, type 2 diabetes mellitus, and if an increase in liver enzymes is accidentally detected against the background of metabolic risk factors, patients should be examined for steatosis, NASH, and fibrosis.

Pathophysiological importance of the microRNA system. Epigenetic factors play an important role in the pathogenesis of NAFLD - changes in gene expression are caused by an adaptive mechanism, and DNA sequence is not associated with changes in gene expression. Among the epigenetic modifications important in NAFLD development, miRNAs are the most studied.
They are short, single-stranded RNA molecules (21-25 nucleotides) that regulate the expression of target gene proteins at the post-transcriptional stage through combination inhibitory mechanisms. [8]
MicroRNAs exhibit various biological functions related to the pathogenesis of NAFLD, including regulation of lipid and glucose metabolism, oxidative stress, endoplasmic reticulum stress, cell differentiation, inflammation, apoptosis, and others. [9]. In experimental models in rats, overexpression of microRNA-34α leads to increased p53 acetylation and activation of p53 through the regulation of sirtuin expression (SIRT 1), which causes hepatocyte apoptosis. [10].

Inhibition of microRNA-34α leads to the activation of transcription factors involved in the body's energy homeostasis: a receptor, peroxisome proliferator-activated protein kinase (PPARα) and 5'AMF-activated protein kinase, which leads to a decrease in lipid content and steatosis in NAFLD rats will come. [11]. MiR-7 regulates fatty acid metabolism through PPARα signaling with lipogenic effects. [12].

In an experimental animal model of NASH, reduction of miR-21 promotes the activation of genes for the development of liver fibrosis: hypoxia-inducible 1-alpha (HIF-1a) and MAP3K protein kinase [13]. MiR-21 has a profibrotic effect, stimulating the SMAD signaling cascade of transforming growth factor (TGFβ), which is the main mediator of fibrogenesis, and the matrix metalloproteinase-2 (MMP2) molecule, which ensures its degradation. [14].

MicroRNA-122 is also involved in the regulation of lipid metabolism. In an experiment on mice, inhibition of microRNA-122 expression led to the stimulation of oxidative processes, a decrease in the amount of fatty acids, and also reduced the level of hepatic steatosis [15].

2. MicroRNA-122 changes in progressive NAFLD. Among liver microRNAs, microRNA-122 is the most abundant [16] and plays a key role in many aspects of liver physiology [17]. The results of human expression studies show that the level of hepatic microRNA-122 increases during the early development of NAFLD, and then gradually decreases in accordance with the development of NASH and fibrosis. Increased microRNA-122 levels have been reported when comparing healthy livers with normal steatosis [18], with decreased levels of microRNA-122 in the livers of obese and NAFLD subjects compared to controls without steatosis.

Circulating levels of microRNA-122 are correlated with alanine aminotransferase (ALT) levels in patients and have higher prognostic and diagnostic value than classical liver function tests, including aspartate aminotransferase (AST) and cytokeratin-18 (CK18) in patients with NAFLD [20, 23]. Inhibition of miR-122 with antisense oligonucleotides in mice reduces hepatic fatty acid and cholesterol synthesis, as well as levels of hepatic fatty acid oxidation, which reduces plasma cholesterol levels [24, 25].

Inhibition of miR-122 with antisense oligonucleotides in mice reduces hepatic fatty acid and cholesterol synthesis, as well as levels of hepatic fatty acid oxidation, which reduces plasma cholesterol. [24, 25]. Saturated fatty acids increase circulating microRNA-122 and decrease its level in hepatocytes, which may result in increased secretion of microRNA-122. [26, 27]

Importance of trace element zinc in non-alcoholic fatty liver disease. Zn trace element is directly related to the development of liver steatosis, it is an important trace element for many enzymes. There is a correlation between daily zinc intake and chronic liver disease, and Zn is essential for the homeostatic function of the liver. Accordingly, deficiency of this mineral impairs liver function, impairs liver tissue regeneration and disease recovery [28].

Deficiency of the micronutrient zinc causes oxidative stress in the liver mitochondria of patients with NASH, which contributes to iron overload, increased insulin resistance, and

the development of liver fibrosis. Oxidative stress plays a key role in the development of NAFLD, especially when the disease progresses from steatosis to steatohepatitis.

The hypothesis that appears in the pathogenesis of nonalcoholic steatohepatitis of the liver is called the "two-hit theory", the first hit is the accumulation of lipids in hepatocytes, and the second hit is oxidative stress. Trace element Zn has also been shown to have a potential effect on reducing lipid peroxidation in an experimental animal model. Because the micronutrient zinc is essential for many oxidants and antioxidants in the body, it is important to remember that Zn deficiency may be associated with obesity through the oxidant-antioxidant system in the liver.

Conclusion

Prognosis and treatment of chronic liver diseases largely depend on the severity of histological changes. Although the biopsy method is the "gold standard" in diagnosis, several of its limitations (invasiveness, subjectivity, i.e., pathological assessment of the process depends on the correct biopsy samples) reduce the frequency of its regular use in clinical practice and encourages the search for new noninvasive and minimally invasive approaches. The main attention is focused on the fact that these methods determine not only the clinical forms of NAFLD, but also have a correlation with the stage of steatosis and fibrosis, the level of morphological activity.

Accumulated data show that microRNAs are important regulators of lipid metabolism. However, each microRNA has multiple targets, and a single gene may be regulated by multiple microRNAs, so it is wrong to evaluate any single microRNA as an independent marker in nonalcoholic fatty liver disease. However, with the increase in the number of new microRNA discoveries, as well as the study of the function of already known specific microRNAs, there is an increasing understanding of the pathogenesis of the disease, the possibilities of using specific microRNAs in clinical practice.

Thus, in the future, defined microRNA profiles, along with biochemical and other laboratory parameters, may form the basis of an integrated scale to assess the stage of liver disease.

References

1. Demir M., Lang S., Steffen H.M. Nonalcoholic fatty liver disease: Current status and future directions. J. Dig. Dis. 2015; 16 (10): 541-57.

2. Lewis J., Mohanty S. Nonalcoholic Fatty Liver Disease: A Review and Update. Dig. Dis. Sci. 2010; 55: 560–78.

3. Sumida Y., Nakajima A., Itoh Y. Limitations of liver biopsy and noninvasive diagnostic tests for the diagnosis of nonalcoholic fatty liver disease/nonalcoholic steatohepatitis. WJG. 2014; 20(2): 475–85.

4. Castera L., Pinzani M. Non-invasive assessment of liver fibrosis: are we ready? Lancet. 2010 Apr 24; 375(9724):1419–20. EASL–EASD–EASO Clinical Practice Guidelines for the management of non-alcoholic fatty liver disease. Journal of Hepatology. 2016; 64:1388–1402.

5. EASL-ALEH Clinical Practice Guidelines: Non-invasive tests for evaluation of liver disease severity and prognosis. Journal of Hepatology. 2015; 63(1): 237 – 64.
6. Kotronen A, Peltonen M, Hakkarainen A, Sevastianova K, Bergholm R, Johansson LM, Lundbom N, Rissanen A, Ridderstråle M, Groop L, Orho–Melander M, Yki–Järvinen H. Prediction of non-alcoholic fatty liver disease and liver fat using metabolic and genetic factors. Gastroenterology. 2009;137(3):865-872. https://doi.org/10.1053/j.gastro.2009.06.005
7. Adams LA, Sanderson S, Lindor KD, Angulo P. The histological course of nonalcoholic fatty liver disease: a longitudinal study of 103 patients with sequential liver biopsies. J Hepatol. 2005;42(1):132-138. https://doi.org/10.1016/j.jhep.2004.09.012
8. Bartel D.P. MicroRNAs: genomics, biogenesis, mechanism, and function. Cell. 2004; 116(2): 281–97.
9. Lakner A.M, Bonkovsky HL, Schrum LW. microRNAs: fad or future of liver disease. WJG. 2011; 17: 2536–42.
10. Castro R.E., Ferreira D.M., Afonso M.B., Borralho P.M., Machado M.V., Cortez-Pinto H., Rodrigues C.M. Mir-34a/SIRT1/p53 is suppressed by ursodeoxycholic acid in the rat liver and activated by disease severity in human non-alcoholic fatty liver disease. J. Hepatol. 2013; 58:119–25.
11. Ding J., Li M., Wan X., Jin X., Chen S., Yu C., Li Y. Effect of mir-34a in regulating steatosis by targeting PPARα expression in nonalcoholic fatty liver disease. Sci. Rep. 2015; 5.
12. Singaravelu R., Quan C.M., Powdrill M.H., Shaw T.A., Srinivasan P., Lyn R.K. et al. MicroRNA-7 mediates cross-talk between metabolic signaling pathways in the liver. Sci Rep. 2018; 8: 36
13. Csak T., Bala S., Lippai D., Satishchandran A., Catalano D., Kodys K. et al. MicroRNA-122 regulates hypoxia-inducible factor-1 and vimentin in hepatocytes and correlates with fibrosis in diet-induced steatohepatitis. Liver Int. 2015; 35: 532–41.
14. Marquez R. T., Bandyopadhyay S., Wendlandt E. B. et al. Correlation between microRNA expression levels and clinical parameters associated with chronic hepatitis C viral infection in humans. Lab. Invest. 2010; 90: 1727—36.
15. Esau C., Davis S., Murray S.F., et al. miR-122 regulation of lipid metabolism revealed by in vivo antisense targeting. Cell Metab. 2006; 3: 87–98. https://doi.org/10.1016/j.jhep.2004.09.012
[16] Hou, J., Lin, L., Zhou, W., Wang, Z., Ding, G., Dong, Q., et al., 2011. Identification of miRNomes in human liver and hepatocellular carcinoma reveals miR-199a/b-3p as therapeutic target for hepatocellular carcinoma. Cancer Cell 19(2):232e243. https://doi.org/10.1016/j.ccr.2011.01.001.
[17] Hu, J., Xu, Y., Hao, J., Wang, S., Li, C., Meng, S., 2012. MiR-122 in hepatic function and liver diseases. Protein Cell 3(5):364e371. https://doi.org/10.1007/s13238-012-2036-3.
[18] Pirola, C.J., Gianotti, T.F., Castaño, G.O., Mallardi, P., Martino, J.S.,

Ledesma, M.M.G.L., et al., 2015. Circulating microRNA signature in nonalcoholic fatty liver disease: from serum non-coding RNAs to liver histology and disease pathogenesis. Gut 64(5):800e812. https://doi.org/10.1136/ gutjnl-2014-306996.
[20] Pirola CJ, Gianotti TF, Castano ˜ GO, Mallardi P, Martino JS, Ledesma MMGL, et al. Circulating microRNA signature in non-alcoholic fatty liver disease: from serum non-coding RNAs to liver histology and disease pathogenesis. Gut 2015, http://dx.doi.org/10.1136/gutjnl-2014-306996.
[21] Thomas M, Deiters A. MicroRNA miR-122 as a therapeutic target for oligonucleotides and small molecules. Curr Med Chem 2013;20:3629–40.
[22] Laudadio I, Manfroid I, Achouri Y, Schmidt D, Wilson MD, Cordi S, et al. A feedback loop between the liver-enriched transcription factor network and Mir-122 controls hepatocyte differentiation. Gastroenterology 2012;142:119–29.
[23] Becker PP, Rau M, Schmitt J, Malsch C, Hammer C, Bantel H, et al. Performance of serum microRNAs -122, -192 and -21 as biomarkers in patients with nonalcoholic steatohepatitis. PLOS ONE 2015;10:e0142661.
[24] Esau C, Davis S, Murray SF, Yu XX, Pandey SK, Pear M, et al. miR-122 regulation of lipid metabolism revealed by in vivo antisense targeting. Cell Metab 2006, http://dx.doi.org/10.1016/j.cmet.2006.01.005.
[25] Krützfeldt J, Rajewsky N, Braich R, Rajeev KG, Tuschl T, Manoharan M, et al. Silencing of microRNAs in vivo with ‘antagomirs’. Nature 2005;438:685–9
[26] Chai C, Rivkin M, Berkovits L, Simerzin A, Zorde-Khvalevsky E, Rosenberg N,et al. Metabolic circuit involving free fatty acids, microRNA 122, and triglyceride synthesis in liver and muscle tissues. Gastroenterology 2017;153:1404–15.
[27] Cheung O, Puri P, Eicken C, Contos MJ, Mirshahi F, Maher JW, et al. Nonalcoholic steatohepatitis is associated with altered hepatic MicroRNA expression. Hepatology 2008;48:1810–20.
28. Kiilerich, S.; Christiansen, C. Distribution of serum zinc between albumin and α2-macroglobulin in patients with different zinc metabolic disorders. Clin. Chim. Acta 1986, 154, 1–6. [CrossRef].