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Review Article

Biochemistry Section

Overview of Genetics in Non Alcoholic Fatty Liver Disease: A Futuristic Cognizance

SANJUKTA MISHRA

ABSTRACT

Non Alcoholic Fatty Liver Disease (NAFLD) is an emerging epidemic worldwide. It comprehends simple steatosis to escalating steatosis with associated fibrosis, cirrhosis, and Hepatocellular Carcinoma (HCC). NAFLD patients are at increased risk of liver-related as well as cardiovascular mortality. It seems to have a robust interconnection with visceral adiposity, Insulin Resistance (IR), inflammation, and environmental factors. Although the pathogenesis of NAFLD is presumed to be linked to lifestyle patterns, nutritional factors, and genetics, the predictor of advancement of this disease spectrum caused by genetics stands imprecise. In the past decade, multiple genome-wide associations and large candidate gene studies have recognised the contribution of several genetic polymorphisms in regulating hepatic lipid metabolism, thus influencing NAFLD establishment and progression. Recent understanding of the genetic underpinning of NAFLD explains the involvement of several common naturally occurring variants in PNPLA3, TM6SF2, MBOAT7, GCKR, LYPLAL1 and PPP1R3B genes in the evolution of the disease. The genetic landscape may play a role to appraise the risk stratification and ascertaining the potential therapeutic target in NAFLD patients. The perspective of this review was to emphasise the genetic basis of the NAFLD spectrum, which modulates the severity and progression of the disease.

Keywords: Cardiovascular, Genetic landscape, Insulin resistance, Steatosis, Therapeutic target

INTRODUCTION

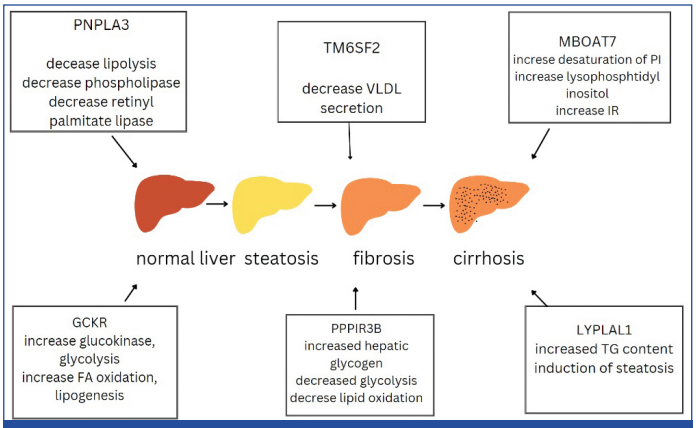
Non Alcoholic Fatty Liver Disease (NAFLD) is a prevailing cause of chronic liver disease globally with a prevalence outlined between 6.7% to 55.1% which accounts for almost one third of the Indian population [1]. It is defined as an enhancement in liver fat content (more than or equal to 5%) due to sources other than imprudent alcohol intake excluding other aetiologies of liver diseases like viral hepatitis, autoimmune liver disease, haemochromatosis, Wilson's disease, and drug-induced liver disease [2]. NAFLD amasses a series of histological varieties extending from elementary fatty liver (steatosis) to Nonalcoholic Steatohepatitis (NASH), which might advance to fibrosis, cirrhosis, and even Hepatocellular Carcinoma (HCC) [3].

Although the molecular backdrop and the intrinsic pathophysiology of NAFLD are not thoroughly acknowledged, the hepatic lipidome is considered a key role player in the development and progress of hepatic steatosis [4]. NAFLD is intimately connected to Metabolic Syndrome (MS) including obesity, hypertension, dyslipidaemia, Insulin Resistance (IR), and Diabetes Mellitus (DM) [5]. It is also conceivable that hyperinsulinaemia-driven de novo lipogenesis and increased influx of free fatty acid may contribute to the esterification and advancement of NAFLD [6]. According to available data, several global surveys of variants across the entire genome or coding region have enlightened the genetic component of NAFLD [7]. Therefore, to better understand this disease, which has become a global public health challenge owing to its increasing prevalence, and chronic hepatic and cardiovascular mortality, it is imperative to understand both environmental and genetic factors. Numerous studies with unique perspectives such as familial aggregation studies, Genome-wide Association Studies (GWAS), and exome-wide association studies have been documented to explain the genetic credibility in NAFLD pathology [7-9].

A more intricate, yet explored outline is the knowledge of heritability assessment according to study design, ethnicity, and methodology used [9]. There is growing corroboration that several common naturally occurring variants in PNPLA3, TM6SF2, MBOAT7, GCKR, PPP1R3B, and LYPLAL1 genes, might regulate hepatic lipid metabolism, thus influencing NAFLD establishment [9,10]. With this background, the present review is an attempt to explore the contemporary evidence on the molecular aspect of NAFLD which might be of use to understanding the genetic risk of this relatively new pandemic disease. That apart, another perspective is to signify the importance of establishing NAFLD diagnosis apropos of chronic liver disease. Besides, the phenomenal improvement in the perception of the genetic risk of NAFLD extends an option to interpret this knowledge into clinical practice. Thus, it may be intended as part of an upcoming personalised management strategy, which is the need of the hour.

GENETIC VARIANTS ASSOCIATED WITH NAFLD

Recent comprehension of the genetic underpinning of NAFLD elucidates the involvement of several naturally occurring variants such as PNPLA3, TM6SF2, MBOAT7, GCKR, LYPLAL1, and PPP1R3B genes in the evolution of the disease [Table/Fig-1] [11,12].

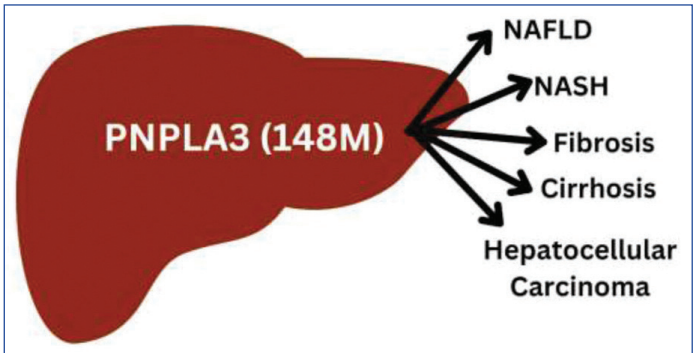


[Table/Fig-1]: Major gene variants associated with NAFLD [11,12].
VLDL: Very low density lipoprotein; IR: Insulin resistance

PNPLA3

There is considerable development of technologies to establish various genetic polymorphisms elaborated in the evolution of

the disease. Several lines of evidence suggest that patatin-like phospholipase domain-containing protein 3 (PNPLA-3, also known as adiponutrin) is a robust genetic determinant of NAFLD [12]. In 2008, the earliest evidence of PNPLA3 being connected to fatty liver conditions was published by GWAS of Hispanic, African, and European American individuals [13]. Subsequently, multiple genetic studies have revealed the association of the PNPLA3 protein isoform 148M variant with a wide spectrum of chronic liver disorders like steatosis, inflammation, fibrosis, and even HCC [Table/Fig-2] [13-16].



[Table/Fig-2]: PNPLA3 linked to wide range of chronic liver diseases [13-16].

Genetic influence of PNPLA3 in the pathogenesis of NAFLD: The magnitude of the genetic influence of PNPLA3 on liver-related outcomes is noteworthy, but still, the underlying pathogenic mechanisms stand elusive. PNPLA3 is located within Hepatocytes and Stellate Cells (HSCs). Its contribution to remodelling lipid droplets is significant. Within hepatocytes, recombinant human PNPLA3 protein isoform 148M binds to lipid droplets. It can hydrolyse Triglycerides (TG) owing to TG lipase activity [17]. Apart from this, it also can facilitate the transfer of Polyunsaturated Fatty Acids (PUFA) from triacyl glycerol and diacylglycerol to phosphocholine [18]. Incidentally, withholding PUFA of TG in the liver due to lower outflow of PUFA into Very Low-Density Lipoprotein (VLDL) TG may alleviate cardiac diseases in PNPAL3 (148M) carriers [18]. PNPLA3 proliferates the fatty load of the liver. However, it is not yet validated to bring on IR [19]. Thus, a beneficial effect of genetic profile may likely be associated with a deleterious consequence on the other part. As mentioned earlier, human PLPA3 is also expressed in HSCs. Within stellate cells, PNPLA3 is regulated by TGF-β to release retinol from retinyl esters. Overexpression of PNPLA3 (148M) leads to HSCs proliferation and chemotaxis. Previous research suggests that c-Jun N-terminal kinase (JNK) is activated to a great extent in PNPLA3 (148M) HSCs and subsequently Peroxisome Proliferator-Activated Receptor protein 1 (PPARγ), a prime HSC quiescence regulator, is inhibited, which adversely controls Liver X Receptor alpha activity (LXRα) [20].

Taken together, these changes might promote cholesterol assemblage in HSCs, which might give rise to fibrogenesis. Hence, PNPLA3 (148M) variants assemblage reorients lipid composition in hepatocytes as well as stellate cells. Taking this into consideration, a therapeutic perspective could be considered, which would directly focus on 148M variants. In a knock-in-mouse (altered gene sequence) model study, PNPLA3 silencing with an Antisense Oligonucleotide (ASO) improves steatohepatitis with remarkable development in the fibrosis stage [21]. Thus, diminishing the mutant PNPLA3 protein would be pertinent from a customised medicine point of view. Further, this study imparts confirmation that PNPLA3 ASO therapy could definitely surpass many hallmark features of NAFLD by repressing the quintessential genetic menace of PNPLA3. It is noteworthy to mention the association of the PNPLA3 genotype with lifestyle intervention. In an ultrasound-based prospective study, it was documented that with stringent calorie restriction, the homozygous 148M/M had a greater reduction of hepatic steatosis compared to 148I/I [22]. Another experiment by Gavril OI et al., proved that PNPLA3 polymorphism was closely connected with hepatic fat content, independent of adiposity or IR [23]. Hence, futuristic therapies associated with PNPLA3 are definitely significant to contemplate.

Transmembrane 6 Superfamily Member 2 (TM6SF2)

NAFLD is a complex disease in which genetic variants and environmental factors interact to determine the disease phenotype. In simpler words, this benign pathological process is highly associated with genetic susceptibility. Transmembrane 6 superfamily member 2 (TM6SF2), another membrane protein located in Golgi, appears to be a potential genetic modifier associated with the development of NAFLD [24]. Pertaining to its polymorphism, the E167K mutation increases the risk of NAFLD by determining the compartmentalisation of lipids within the intracellular droplet. A recent exome-wide association study could identify a prospective association between TM6SF2 polymorphism and hepatic TG accumulation [25]. Apart from this, a few functional studies also confirmed the crucial role of TM6SF2 (E 167K variant) in VLDL secretion [26,27]. Thus, briefly, it is speculated that the TM6SF2 E167K variant results in an enhancement of hepatic TG by lowering VLDL secretion, and also it is verified to be associated with the illustration of other lipid-related genes (PNPLA3, ACSS2, and others) [26]. Yet the exact mechanism remains to be determined. Intriguingly, the connection between the TM6SF2 E167K variant and hepatic TG is unconnected to the outcome of the PNPLA3 I148M variant [28].

In the light of these findings, various studies have explored the significant association between genetic variation in transmembrane 2 and histological disease severity in NAFLD [29-31]. Hepatic fat content in the analysed patients was significantly higher in TM6SF2 carriers. Sookoian S et al., perceived a significant association between TM6SF2 and the degree of hepatic steatosis [32]. In agreement with the above study, one more study conducted on NAFLD patients of different stages documented the association between TM6SF2 variant and the risk of steatosis [29]. Earlier, a link between TM6SF2 genetic variant and the severity of steatosis had already been established [24]. Several other studies also explored the above genetic association with the severity of the histological form of NAFLD [29,33]. Based on available findings, the TM6SF2 E167K variant also gives out to high susceptibility to NAFLD-related HCC without the association of risk factors like age, gender, and diabetes [32]. Moreover, a strong interplay between TM6SF2 and increased risk of NAFLD and decreased risk of cardiac disease remains to be determined. Thus, a better understanding of the biological function of this enigmatic protein will facilitate a therapeutic and preventive strategy for NAFLD in high-risk populations.

MBOAT7

A multispanning transmembrane protein, Membrane-Bound O-Acyltransferase Domain containing 7 (MBOAT7) documented to be one among the genes associated with NAFLD, encodes Lysophosphatidylinositol Acyltransferase 1 (LPIAT1) [34]. It catalyses the transfer of PUFA arachidonyl CoA to lysophosphatidyl inositol, thus helping in the remodelling and desaturation of 2nd acyl chain of phospholipid [34]. It is acknowledged that MBOAT7 has been attested to human hepatocytes, sinusoidal endothelial cells, and HSCs [35]. Notably, it acts as an effectual stimulator for hepatic inflammation and fibrosis owing to its reformation to eicosanoids [36]. The recognised human risk variant MBOAT7 rs641738T is of great interest because of its association with NAFLD [37-40]. Furthermore, it has been documented that loss of hepatic MBOAT7 might lead to liver fibrosis [41]. Though several lines of evidence denote histopathological correlations, still the mechanism by which the rs64138T variant ushers to NAFLD development and hepatic fibrosis, is not extensively appreciated. Functional studies

line up to interpret the process through which genetic variations influence the outcome of fatty liver disease [40-43]. Luukkonen PK et al., observed reduced levels of MBOAT7 protein in the liver and hepatic remodelling in carriers of rs641738T [40]. Mancina RM et al., confirmed the association of rs641738 variant with steatosis severity of NAFLD in European descent [38]. Furthermore, it has been established that MBOAT7 is linked to IR as MBOAT7 suppression is observed during obesity, which prompts NAFLD [42]. Another piece of evidence by Sookoian S et al., corroborates the finding that MBOAT7 is down-regulated in NAFLD patients even independently of the presence of the rs641738 polymorphism [43]. Notably, the causative role of MBOAT7 has been further confirmed by Meroni M et al., who illustrated hepatic fat accumulation by acute silencing of hepatic MBOAT7 [35].

GCKR

Considering the differences in the genetic background of NAFLD patients, it is essential to recognise the association between gene polymorphisms and the progression of NAFLD. Glucokinase Regulatory protein (GCKR) gene polymorphism seems to be an important validated risk gene in the development of NAFLD [44]. GCKR codes for glucokinase regulatory protein which gets attached to the phosphorylating glucokinase enzyme that controls hepatic glucose metabolism and lipogenesis [45]. Several GWAS ventured to interpret the involvement of GCKR gene polymorphism in predisposition to NAFLD. But the results have been inconsistent and contentious [46-49]. GWAS by Speliotes EK et al., reported a strong association between GCKR rs 780094 and NAFLD risk [46]. The study by Tan HL et al., was in complete agreement with the above result showing a similar significant genetic association [47]. Conversely, several community-based studies could find no such strong linkage, which aims toward the need for additional assessment [48,49]. Accumulated studies also focus to decode the congruity between GCKR rs 1260326 with the risk of NAFLD. The meta-analysis by Li J et al., suggested a significant association between GCKR rs 1260326 and increased risk of NAFLD [50]. These contemporary results furnish an inventive perception of GCKR function which possibly helps in leading the focus for further studies.

PPP1R3B

The genetic variants in protein modulating hepatocellular lipid handling function as a prognostic factor. GWAS could identify another genetic locus PPP1R3B (Protein phosphatase 1 regulatory subunit 3B), which has been documented to be associated with hepatic fat content [51]. PPP1R3B encodes a protein that promotes hepatic glycogen synthesis. Furthermore, it has been hypothesised that rs4240624 variant is related to reduced hepatic fat content and increased glycogen content [52]. Even with few contrasting data, work by Dongiovanni P et al., provide convincing support for the fact that PPP1R3B protects against hepatic fat accumulation [52].

Increased hepatic glycogen content has prompted the likelihood of shunting glucose from glycolysis to glycogenesis. It could also be speculated that there will be suppression of de novo lipogenesis. Notably reduced glycolysis and lipid oxidation might lead to reduced activation of the inflammatory and fibrogenic pathways [52]. Stender S et al., noted the association of PPP1R3B with a benign form of hepatic steatosis, which was consistent with the notion that it promotes a mild form of hepatic glycogenesis [51]. Overall, in addition to raised calorie intake, genetic and epigenetic liability promotes the development of NAFLD [53]. Further clinical research might recommend enlightenment on the involvement of specific Single Nucleotide Polymorphisms (SNPs) in the progression of NAFLD. Interpreting the connection between fatty liver and changes in circulating lipid metabolite is useful in achieving new possibilities for the treatment and prevention of this complex disease.

LYPLAL1

The metabolic association profile of genotype highlights the heterogeneity of molecular pathways associated with fatty liver. In the last few years, GWAS has come up with convincing evidence about the genetic variant association of NAFLD. Lysophospholipase-like 1 (LYPLAL1) is one of the few important genetic sequence variants, which contribute to the pathogenesis of NAFLD [54]. LYPLAL1 protein, linked to the human body's fat distribution, might act as TG lipase in adipose tissues. The relevance of SNP LYPLAL1 has been identified in multiple ethnic groups by recent GWAS [54,55]. Among these variants, the function of LYPLAL1 rs12137855-C is documented to be linked to Computed Tomography (CT)-defined steatosis and biopsy-evinced NAFLD in end-stage fibrosis patients in candidate gene resequencing strategy [56] Multivariable analysis in the East Asian population confirmed that the metabolic effects of LYPLAL1 rs12137855-C were similar but statistically less robust, which could be attributed to the limited sample size [55]. Definitely, research to a greater extent on this point is imperative hereafter. Findings in previous studies are tabulated in [Table/Fig-3] [15,17-19,21-23,25-27,29-31,37-45,47-52,54-55].

Genetic variants	Author	Ref. No.	Country and year of study	Findings of study
PNPLA3	Gavril OI et al.,	[23]	Romania, 2021	no correlation between PNPLA3 and NAFLD type 2 DM
	Luukkonen PK et al.,	[18]	Finland, 2019	Remodels liver triglyceride in a polyunsaturated direction
	Basu Ray S et al.,	[17]	USA, 2019	Accumulation of PNPLA3 on hepatic lipid droplet
	Franko A et al.,	[19]	Germany, 2018	No impairment in insulin sensitivity in PNPLA3 carrier
	Linden D et al.,	[21]	Sweden, 2019	PNPLA3 silencing ameliorates Nonalcoholic Steatohepatitis (NASH) and fibrosis
	Krawczyk M et al.,	[22]	Germany, 2016	Effect of calorie restriction on liver phenotype
	Shen J et al.,	[15]	Hong Kong, 2015	PNPLA3 genotype confers a higher risk of NAFLD
TM6SF2	Kozlitina J et al.,	[25]	USA, 2014	Impaired TM6SF2 activity contributes to impaired VLDL secretion and NAFLD
	Krawczyk M et al.,	[37]	Germany, 2017	Modulate hepatic fat accumulation which influences NAFLD severity
	Wang X et al.,	[30]	China, 2015	Increased NAFLD risk in Chinese population independent of other genetic factors
	Zhou Y et al.,	[31]	Finland, 2015	Lowering of circulating triglyceride
	Mahdessian H et al.,	[26]	Sweden, 2014	Regulator of liver fat metabolism with opposing effect on the secretion of triglyceride-rich lipoprotein
	Holmen OL et al.,	[27]	Norway, 2014	Effect on total cholesterol level
	Liu YL et al.,	[29]	UK, 2014	Influence hepatic fibrosis progression
MBOAT 7	Mancina RM et al.,	[38]	Sweden, 2016	Association with NAFLD and steatosis severity in European descent
	Di Sessa A et al.,	[39]	Italy, 2018	Pediatric association with markers of liver fibrosis
	Sookoian S et al.,	[43]	Argentina, 2018	Down-regulation of genetic factor in NAFLD
	Thangapandi VR et al.,	[41]	Germany, 2021	Deficiency leads to liver fibrosis via lipid signalling
	Helsley RN et al.,	[42]	USA, 2019	Link with Insulin Resistance (IR) that prompts NAFLD
	Luukkonen PK et al.,	[40]	Finland, 2016	Increases severity by altering hepatic phosphatidyl inositol

GCKR	Li J et al.,	[50]	China, 2021	Meta-analysis confirming the role in NAFLD
	Gao H et al.,	[44]	China, 2019	Association with risk of NAFLD and coronary artery disease in the Chinese population
	Tan HL et al.,	[47]	Malaysia, 2014	Association found with risk and severity
	Peter A et al.,	[45]	Germany, 2011	Increased activity induce fatty liver and its metabolic consequences
	Yang H et al.,	[48]	China, 2018	Combination with other genetic polymorphisms showing better diagnostic accuracy
PPP1R3B	Dongiovanni P et al.,	[52]	Italy, 2018	Protection against fat accumulation and fibrosis in individuals at high risk
	Stender S et al.,	[51]	USA, 2018	Association with mild hepatic glycogenesis causing liver injury
LYPLAL1	Sliz E et al.,	[54]	Finland, 2018	Increased concentration of circulating lipid
	Wang X et al.,	[49]	China, 2016	No significant association in the Han Chinese population
	Di Costanzo A et al.,	[55]	Italy, 2018	Evaluation of polygenic determinant

[Table/Fig-3]: Genetic variants associated with NAFLD [15,17-19,21-23,25-27,29-31,37-45,47-52,54,55].

CONCLUSION(S)

Strikingly in recent years, genetic association studies have accentuated the role of genetic components as general modulators of NAFLD and increasing evidence supports the role of SNPs in the risk and development of NAFLD. Genetic factors associated with lipid biology (PNPLA3, TM6SF2, MBOAT7, GCKR, PPP1R3B, and LYPLAL1) may impact disease development and progression. Though the exact pathogenesis is not yet clarified completely, increasing evidence supports the complex interplay between the individual and cumulative contributions of identified genes. Forthwith, genetics is a domain of considerable interest as it could anticipate new insights into therapeutic targets and non-invasive biomarkers. Definite and detailed genetic expertise attained with explanatory techniques should enable the medical fraternity to strengthen precision medicine with reference to NAFLD. Thus, it may be considered as part of the forthcoming customised management strategy, which is an absolute necessity.

REFERENCES

[1] Shalimar, Elhence A, Bansal B, Gupta H, Anand A, Singh TP, et al. Prevalence of non-alcoholic fatty liver disease in india: A systematic review and meta-analysis. J Clin Exp Hepatol. 2022;12(3):818-29. Doi: 10.1016/j.jceh.2021.11.010.

[2] Chalasani N, Younossi Z, Lavine JE, Charlton M, Cusi K, Rinella M, et al. 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):328-57. Doi.org/10.1002/hep.29367.

[3] Pouwels S, Sakran N, Graham Y, Leal A, Pintar T, Yang W, et al. Non-alcoholic fatty liver disease (NAFLD): A review of pathophysiology, clinical management and effects of weight loss. BMC Endocr Disord. 2022;22(1):63. Doi.org/10.1186/s12902-022-00980-1.

[4] Wang J, Su Z, Feng Y, Xi R, Liu J, Wang P. Comparison of several blood lipid-related indexes in the screening of non-alcoholic fatty liver disease in women: A cross-sectional study in the Pearl River Delta region of southern China. BMC Gastroenterol. 2021;21(1):482. Doi.org/10.1186/s12876-021-02072-1.

[5] Godoy-Matos AF, Silva Júnior WS, Valerio CM. NAFLD as a continuum: From obesity to metabolic syndrome and diabetes. Diabetol Metab Syndr. 2020;12:60. Doi.org/10.1186/s13098-020-00570-y.

[6] Vvedenskaya O, Rose TD, Knittelfelder O, Palladini A, Wodke JAH, Schuhmann K, et al. Nonalcoholic fatty liver disease stratification by liver lipidomics. J Lipid Res. 2021;62:100104. Doi: 10.1016/j.jlr.2021.100104.

[7] Eslam M, Valenti L, Romeo S. Genetics and epigenetics of NAFLD and NASH: Clinical impact. J Hepatol. 2018;68(2):268-79. Doi: 10.1016/j.jhep.2017.09.003.

[8] Sookoian S, Pirola CJ. Genetic predisposition in nonalcoholic fatty liver disease. Clin Mol Hepatol. 2017;23(1):01-12. Doi: 10.3350/cmh.2016.0109.

[9] Chandrasekharan K, Alazawi W. Genetics of non-alcoholic fatty liver and cardiovascular disease: Implications for therapy? Front Pharmacol. 2020;10:1413. Doi: 10.3389/fphar.2019.01413.

[10] Eslam M, George J. Genetic contributions to NAFLD: Leveraging shared genetics to uncover systems biology. Nat Rev Gastroenterol Hepatol. 2020;17(1):40-52. Doi: 10.1038/s41575-019-0212-0.

[11] Sookoian S, Pirola CJ, Valenti L, Davidson NO. Genetic pathways in nonalcoholic fatty liver disease: Insights from systems biology. Hepatology. 2020;72:330-46. Doi: 10.1002/hep.31229.

[12] Yuan L, Terrault NA. PNPLA3 and nonalcoholic fatty liver disease: Towards personalized medicine for fatty liver. Hepatobiliary Surg Nutr. 2020;9(3):353-56. Doi: 10.21037/hbsn.2019.10.35.

[13] Dong XC. PNPLA3-A potential therapeutic target for personalized treatment of chronic liver disease. Front Med (Lausanne). 2019;6:304. Doi: 10.3389/fmed.2019.00304.

[14] Salameh H, Raff E, Erwin A, Seth D, Nischalke HD, Falleti E, et al. PNPLA3 gene polymorphism is associated with predisposition to and severity of alcoholic liver disease. Am J Gastroenterol. 2015;110(6):846-56. Doi: 10.1038/ajg.2015.137.

[15] Shen J, Wong GL, Chan HL, Chan RS, Chan HY, Chu WC, et al. PNPLA3 gene polymorphism and response to lifestyle modification in patients with nonalcoholic fatty liver disease. J Gastroenterol Hepatol. 2015;30(1):139-46. Doi: 10.1111/jgh.12656.

[16] Stickel F, Buch S, Nischalke HD, Weiss KH, Gotthardt D, Fischer J, et al. Genetic variants in PNPLA3 and TM6SF2 predispose to the development of hepatocellular carcinoma in individuals with alcohol-related cirrhosis. Am J Gastroenterol. 2018;113(10):1475-83. Doi: 10.1038/s41395-018-0041-8.

[17] BasuRay S, Wang Y, Smagris E, Cohen JC, Hobbs HH. Accumulation of PNPLA3 on lipid droplets is the basis of associated hepatic steatosis. Proc Natl Acad Sci USA. 2019;116(19):9521-26. Doi.org/10.1073/pnas.1901974116.

[18] Luukkonen PK, Nick A, Hölttä-Vuori M, Thiele C, Isokuortti E, Lallukka-Brück S, et al. Human PNPLA3-I148M variant increases hepatic retention of polyunsaturated fatty acids. JCI Insight. 2019;4(16):e127902. Doi: 10.1172/jci.insight.127902.

[19] Franko A, Merkel D, Kovarova M, Hoene M, Jaghutriz BA, Heni M, et al. Dissociation of fatty liver and insulin resistance in I148M PNPLA3 carriers: Differences in Diacylglycerol (DAG) FA18:1 lipid species as a possible explanation. Nutrients. 2018;10(9):1314. Doi: 10.3390/nu10091314.

[20] Bruschi FV, Claudel T, Tardelli M, Caligiuri A, Stulnig TM, Marra F, et al. The PNPLA3 I148M variant modulates the fibrogenic phenotype of human hepatic stellate cells. Hepatology. 2017;65(6):1875-90. Doi: 10.1002/hep.29041.

[21] Lindén D, Ahnmark A, Pingitore P, Ciociola E, Ahlstedt I, Andréasson AC, et al. Pnpla3 silencing with antisense oligonucleotides ameliorates nonalcoholic steatohepatitis and fibrosis in Pnpla3 I148M knock-in mice. Mol Metab. 2019;22:49-61. Doi: 10.1016/j.molmet.2019.01.013.

[22] Krawczyk M, Stachowska E, Milkiewicz P, Lammert F, Milkiewicz M. Reduction of caloric intake might override the prosteatotic effects of the PNPLA3 p.I148M and TM6SF2 p.E167K variants in patients with fatty liver: Ultrasound-based prospective study. Digestion. 2016;93(2):139-48. Doi: 10.1159/000441185.

[23] Gavril OI, Arhire LI, Gavril RS, Zota MI, Gherasim A, Nita O, et al. Correlations between PNPLA3 gene polymorphisms and NAFLD in type 2 diabetic patients. Medicina (Kaunas). 2021;57(11):1249. Doi: 10.3390/medicina57111249.

[24] Chen LZ, Xia HH, Xin YN, Lin ZH, Xuan SY. TM6SF2 E167K variant, a novel genetic susceptibility variant, contributing to nonalcoholic fatty liver disease. J Clin Transl Hepatol. 2015;3(4):265-70. Doi: 10.14218/JCTH.2015.00023.

[25] Kozlitina J, Smagris E, Stender S, Nordestgaard BG, Zhou HH, Tybjærg-Hansen A, et al. Exome-wide association study identifies a TM6SF2 variant that confers susceptibility to nonalcoholic fatty liver disease. Nat Genet. 2014;46(4):352-56. Doi: 10.1038/ng.2901.

[26] Mahdessian H, Taxiarchis A, Popov S, Silveira A, Franco-Cereceda A, Hamsten A, et al. TM6SF2 is a regulator of liver fat metabolism influencing triglyceride secretion and hepatic lipid droplet content. Proc Natl Acad Sci U S A. 2014;111(24):8913-28. Doi: 10.1073/pnas.1323785111.

[27] Holmen OL, Zhang H, Fan Y, Hovelson DH, Schmidt EM, Zhou W, et al. Systematic evaluation of coding variation identifies a candidate causal variant in TM6SF2 influencing total cholesterol and myocardial infarction risk. Nat Genet. 2014;46(4):345-51. Doi: 10.1038/ng.2926.

[28] Ruhanen H, Perttilä J, Hölttä-Vuori M, Zhou Y, Yki-Järvinen H, Ikonen E, et al. PNPLA3 mediates hepatocyte triacylglycerol remodeling. J Lipid Res. 2014;55(4):739-46. Doi: 10.1194/jlr.M046607.

[29] Liu YL, Reeves HL, Burt AD, Tiniakos D, McPherson S, Leathart JB, et al. TM6SF2 rs58542926 influences hepatic fibrosis progression in patients with non-alcoholic fatty liver disease. Nat Commun. 2014;5:4309. Doi: 10.1038/ncomms5309.

[30] Wang X, Liu Z, Peng Z, Liu W. The TM6SF2 rs58542926 T allele is significantly associated with non-alcoholic fatty liver disease in Chinese. J Hepatol. 2015;62(6):1438-39. Doi: 10.1016/j.jhep.2015.01.040.

[31] Zhou Y, Llauradó G, Orešič M, Hyötyläinen T, Orho-Melander M, Yki-Järvinen H. Circulating triacylglycerol signatures and insulin sensitivity in NAFLD associated with the E167K variant in TM6SF2. J Hepatol. 2015;62:657-63. Doi: 10.1016/j.jhep.2014.10.010.

[32] Sookoian S, Castaño GO, Scian R, Mallardi P, Fernández Gianotti T, Burgueño AL, et al. Genetic variation in transmembrane 6 superfamily member 2 and the risk of non-alcoholic fatty liver disease and histological disease severity. Hepatology. 2015;61:515-25. Doi: 10.1002/hep.27556.

[33] Dongiovanni P, Petta S, Maglio C, Fracanzani AL, Pipitone R, Mozzi E, et al. Transmembrane 6 superfamily member 2 gene variant disentangles nonalcoholic steatohepatitis from cardiovascular disease. Hepatology. 2015;61(2):506-14. Doi: 10.1002/hep.27490.

[34] Teo K, Abeysekera KWM, Adams L, Aigner E, Anstee QM, Banales JM, et al. rs641738C>T near MBOAT7 is associated with liver fat, ALT and fibrosis in NAFLD: A meta-analysis. J Hepatol. 2021;74(1):20-30. Doi: 10.1016/j.jhep.2020.08.027.

[35] Meroni M, Longo M, Fracanzani AL, Dongiovanni P. MBOAT7 down-regulation by genetic and environmental factors predisposes to MAFLD. E Bio Medicine. 2020;57:102866. Doi: 10.1016/j.ebiom.2020.102866.

[36] Zarini S, Hankin JA, Murphy RC, Gijón MA. Lysophospholipid acyltransferases and eicosanoid biosynthesis in zebrafish myeloid cells. Prostaglandins Other Lipid Mediat. 2014;113-115:52-61. Doi: 10.1016/j.prostaglandins.2014.08.003.
[37] Krawczyk M, Rau M, Schattenberg JM, Bantel H, Pathil A, Demir M, et al. NAFLD clinical study group. Combined effects of the PNPLA3 rs738409, TM6SF2 rs58542926, and MBOAT7 rs641738 variants on NAFLD severity: A multicenter biopsy-based study. J Lipid Res. 2017;58(1):247-55. Doi: 10.1194/jlr.P067454.
[38] Mancina RM, Dongiovanni P, Petta S, Pingitore P, Meroni M, Rametta R, et al. The MBOAT7-TMC4 variant rs641738 increases risk of nonalcoholic fatty liver disease in individuals of European Descent. Gastroenterology. 2016;150(5):1219-30.e6. Doi: 10.1053/j.gastro.2016.01.032.
[39] Di Sessa A, Umano GR, Cirillo G, Del Prete A, Iacomino R, Marzuillo P, et al. The membrane-bound O-Acyltransferase7 rs641738 variant in pediatric nonalcoholic fatty liver disease. J Pediatr Gastroenterol Nutr. 2018;67(1):69-74. Doi: 10.1097/MPG.0000000000001979.
[40] Luukkonen PK, Zhou Y, Hyötyläinen T, Leivonen M, Arola J, Orho-Melander M, et al. The MBOAT7 variant rs641738 alters hepatic phosphatidylinositols and increases severity of non-alcoholic fatty liver disease in humans. J Hepatol. 2016;65(6):1263-65. Doi: 10.1016/j.jhep.2016.07.045.
[41] Thangapandi VR, Knittelfelder O, Brosch M, Patsenker E, Vvedenskaya O, Buch S, et al. Loss of hepatic Mboat7 leads to liver fibrosis. Gut. 2021;70:940-50.
[42] Helsley RN, Varadharajan V, Brown AL, Gromovsky AD, Schugar RC, Ramachandiran I, et al. Obesity linked suppression of membrane-bound O-acyltransferase 7 (MBOAT7) drives non-alcoholic fatty liver disease. E-life. 2019;8:e49882. Doi: 10.7554/eLife.49882.
[43] Sookoian S, Flichman D, Garaycoechea ME, Gazzi C, Martino JS, Castaño GO, et al. Lack of evidence supporting a role of TMC4-rs641738 missense variant MBOAT7-intergenic downstream variant-in the susceptibility to nonalcoholic fatty liver disease. Sci Rep. 2018;8(1):5097. Doi: 10.1038/s41598-018-23453-9.
[44] Gao H, Liu S, Zhao Z, Yu X, Liu Q, Xin Y, et al. Association of GCKR gene polymorphisms with the risk of nonalcoholic fatty liver disease and coronary artery disease in a Chinese Northern Han population. J Clin Transl Hepatol. 2019;7(4):297-303. Doi: 10.14218/JCTH.2019.00030.
[45] Peter A, Stefan N, Cegan A, Walenta M, Wagner S, Königsrainer A, et al. Hepatic glucokinase expression is associated with lipogenesis and fatty liver in humans. J Clin Endocrinol Metab. 2011;96(7):E1126-30. Doi: 10.1210/jc.2010-2017.
[46] Speliotes EK, Yerges-Armstrong LM, Wu J, Hernaez R, Kim LJ, Palmer CD, et al. Genomewide association analysis identifies variants associated with nonalcoholic fatty liver disease that have distinct effects on metabolic traits. PLoS Genet. 2011;7(3):e1001324. Doi: 10.1371/journal.pgen.1001324.
[47] Tan HL, Zain SM, Mohamed R, Rampal S, Chin KF, Basu RC, et al. Association of glucokinase regulatory gene polymorphisms with risk and severity of non-alcoholic fatty liver disease: An interaction study with adiponutrin gene. J Gastroenterol. 2014;49(6):1056-64. Doi: 10.1007/s00535- 013-0850-x
[48] Yang H, Chen G, Song C, Li D, Ma Q, Chen G, et al. A novel index including SNPs for the screening of nonalcoholic fatty liver disease among elder Chinese: A population-based study. Medicine (Baltimore). 2018;97(13):e0272. Doi: 10.1097/MD.0000000000010272.
[49] Wang X, Liu Z, Wang K, Wang Z, Sun X, Zhong L, et al. Additive effect of risk alleles of PNPLA3 and TM6SF2 on non-alcoholic fatty liver disease (NAFLD) in a Chinese population. Front Genet. 2016;7:140. Doi: 10.3389/fgene.2016.00140.
[50] Li J, Zhao Y, Zhang H, Hua W, Jiao W, Du X, et al. Contribution of Rs780094 and Rs1260326 polymorphisms in GCKR gene to nonalcoholic fatty liver disease: A meta-analysis involving 26,552 participants. Endocr Metab Immune Disord Drug Targets. 2021;21(9):1696-708. Doi: 10.2174/1871530320999201126202706.
[51] Stender S, Smagris E, Lauridsen BK, Kofoed KF, Nordestgaard BG, Tybjaerg-Hansen A, et al. Relationship between genetic variation at PPP1R3B and levels of liver glycogen and triglyceride. Hepatology. 2018;67(6):2182-95. Doi: 10.1002/hep.29751.
[52] Dongiovanni P, Meroni M, Mancina RM, Baselli G, Rametta R, Pelusi S, et al. Protein phosphatase 1 regulatory subunit 3B gene variation protects against hepatic fat accumulation and fibrosis in individuals at high risk of nonalcoholic fatty liver disease. Hepatol Commun. 2018;2(6):666-75. Doi: 10.1002/hep4.1192.
[53] Jonas W, Schürmann A. Genetic and epigenetic factors determining NAFLD risk. Mol Metab. 2021;50:101111. Doi: 10.1016/j.molmet.2020.101111.
[54] Sliz E, Sebert S, Würtz P, Kangas AJ, Soininen P, Lehtimäki T, et al. NAFLD risk alleles in PNPLA3, TM6SF2, GCKR and LYPLAL1 show divergent metabolic effects. Hum Mol Genet. 2018;27(12):2214-23. Doi: 10.1093/hmg/ddy124.
[55] Di Costanzo A, Belardinilli F, Bailetti D, Sponziello M, D'Erasmo L, Polimeni L, et al., Evaluation of polygenic determinants of Nonalcoholic Fatty Liver Disease (NAFLD) by a candidate genes resequencing strategy. Sci Rep. 2018;8(1):3702. Doi: 10.1038/s41598-018-21939-0.

PARTICULARS OF CONTRIBUTORS:
1. Professor, Department of Biochemistry, Kalinga Institute of Medical Science, KIIT University, Bhubaneswar, Odisha, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Prof. Sanjukta Mishra,
Department of Biochemistry, Kalinga Institute of Medical Science, KIIT University, Bhubaneswar, Odisha, India.
E-mail: drmsanjukta@gmail.com

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