ISSN: 0975-833X

Available online at http://www.journalcra.com

International Journal of Current Research

Vol. 16, Issue, 10, pp.30278-30282, October, 2024
DOI: https://doi.org/10.24941/ijcr.47944.09.2024

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

RESEARCH ARTICLE

POSSIBLE GENETIC INVOLVEMENT IN NON - ALCOHOLIC FATTY LIVER DISEASE

Dalmia[1], Deeksha[1] and Dr. Paras Monga[1,2]

[1]Department of Pharmacy Practice, CT Institute of Pharmaceutical Sciences, CT Group of Institutions, Jalandhar, 144024, Punjab, India; [2]Department of Pharmaceutical Sciences, M M College of Pharmacy, MMDU Mullana, Ambala- Haryana

ARTICLE INFO

Article History:
Received 24th July, 2024
Received in revised form
17th August, 2024
Accepted 29th September, 2024
Published online 30th October, 2024

Key Words:
Non - Alcoholic Fatty Liver Disease (NAFLD); Genome – Wide Association Studies (GWAS);Paptatin-Like Phospholipase Domain Containing Protein 3(PNPLA3);Transmembrane 6 Superfamily Member 2 (TM6SF2);Membrane Bound O-Acyltransferase Domain Containing 7(MBOAT7); Superoxide Dismutase 2 (SOD2).

ABSTRACT

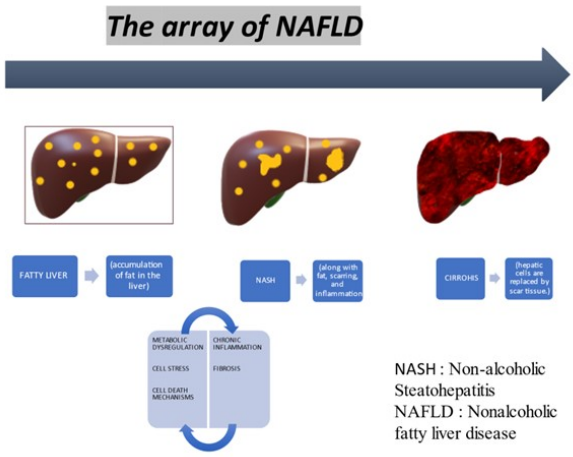
The most common condition affecting the liver is non-alcoholic liver disease (NAFLD). It is well known that the majority of people who suffer from nonalcoholic fatty liver disease (NAFLD) have one or more of the recently identified metabolic syndrome symptoms, including central obesity, impaired glucose tolerance, hypertension, and dyslipidemia. NAFLD is a group of illnesses that ranges from simple steatosis, which is defined by hepatocyte damage, inflammation, and a fatty liver, to steatohepatitis, fibrosis, and cirrhosis. NAFLD is a complicated disease. Through the promotion of liver inflammation, cell necrosis and apoptosis, and the creation of fibrosis, Cytokines may have a direct role in the development of nonalcoholic fatty liver disease (NAFLD). Adiponectin levels rise in NASH patients receiving thiazolidinedione therapy, indicating improvement in hepatic steatosis, necroinflammation, and—most importantly—fibrosis.Obese patients who inherit both high TGF-1 and angiotensinogen -producing polymorphisms may be more susceptible to advanced fibrosis.There have been a number of candidate gene studies and a small number of GWAS (genome-wide association studies) in NAFLD populations as a result of the heritability and complexity of NAFLD.PNPLA3 was the first variant associated with NAFLD.Recent GWA studies have demonstrated a relationship between the SNP rs58542926 C>T in theTM6SF2 gene and the severity of NAFLD. Both raised serum triglyceride levels and an enhanced fibrosis risk in NAFLD patients are linked to the GCKR P446L variant.An increase in inflammation and hepatic fibrosis in NAFLD patients is linked to the MBOAT7 variant.

Citation: Dalmia, Deeksha and Dr. Paras Monga. 2024. "Possible genetic involvement in non - alcoholic fatty liver disease'.*". International Journal of Current Research*, 16, (10), 30278-30282.

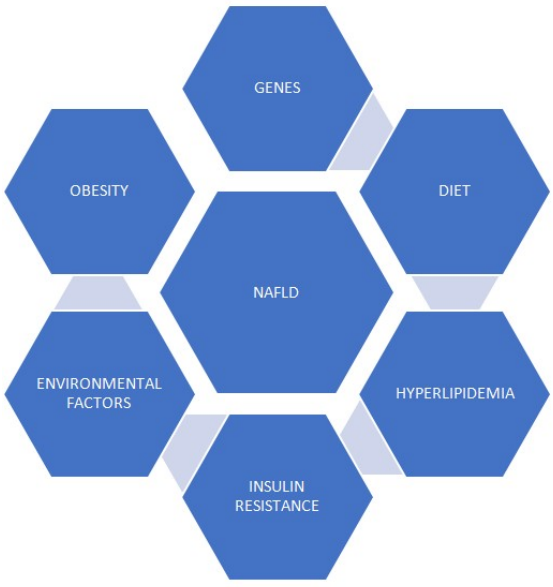
INTRODUCTION

The most frequent condition affecting the liver is non-alcoholic liver disease (NAFLD).NAFLD is expanding rapidly to become an all-around health crisis. Infact, it is the most prevalent liver condition both domestically and internationally. As the most widespread type of live disease worldwide, NAFLD prevalence keeps rising. It is widely accepted that the majority of individuals withnon-alcoholic fatty liver disease exhibit one or more identified metabolic syndrome symptoms, including obesity, impaired glucose tolerance, hypertension, and hyperdyslipemia. Non-alcoholic Steatohepatitis (NASH) and fibrosis are more advanced diseases among these individuals; the number of metabolic characteristics present andthe severity of the underlying insulin resistance are interconnected with these conditions[i]. However, the majority of patients who have multiple metabolic syndrome symptoms will only have simple steatosis.Althoughthe pathophysiology of NAFLD is complicated, increasing visceral adiposity, enhanced insulin resistance, and free fatty acid release possess a crucial role in the early development and maintenance of hepatic steatosis.

Infact, NAFLD is often regarded as a hepatic symptomof generalized insulin resistance [ii]. In this situation, neutral fat builds up. In hepatocytes' internal lipid droplets, primarily under the Several lipid substances are involved, but they take the form of triglycerides. Triglyceride esterification serves as a protective mechanismfor fat storage that is driven by hyperinsulinemia and derived from insulin-resistant and inflammatory adipose tissue, diet, de novo lipogenesis, etc. In fact, if free fatty acids weren't present, they would cause lipotoxicity and activate fibrogenesis, a liver condition [iii]. The metabolism of lipid droplets is a tightly controlledprocess, as a number of proteins implicated in the liver's pathophysiology are damaged [iv]. NAFLD is an array of diseases thatincludes steatohepatitis, fibrosis, and cirrhosis, together with simple steatosis, which is known to be caused by inflammation, afatty liver, and damage to hepatic cells [v].Drugs and toxins are also inducers of NAFLD. It is clear that NAFLD is a complex disorder with many interacting metabolic pathwaysthat appear to be regulatedby combining the effects of environment-related factors and genetic redistribution. Among these, nutrient abundance is the most crucial one, particularly in light of the normal Western diet's emphasis on simple carbohydrates,



saturated fat, and highly processed foods. This can lead to a calorie imbalance and increased weight gain in most people when combined with a Western sedentary lifestyle. A higher prevalence of overweight or obesity is linked to a higher incidence of NAFLD in both adult and pediatric populations [vi].Several variations in genes associated with NAFLD or NASH have been discovered by candidate gene strategies and genome-wide association studies (GWAS) [vii].



In this review, we concentrated on the genetic component of NAFLD, with a focus on the function of genetics in the etiology and natural history of the illness. The genetic vulnerability among lean people with NAFLD and the potential utility of genetic testing in identifying those at risk are also discussed in detail [viii].

Pathogenesis of NAFLD: NAFLD is a complicated disorder. The development of steatosis, hepatic inflammatory responses, and fibrogenesis can all be influenced by a variety of parallel or sequential, hierarchically arranged, and illness-related events [ix]. In NASH, Kupffer cells may create cytokines, particularly tumor necrosis factor α (TNF-α), in reaction to gut-derived endotoxin. On the other hand, hepatocytes or adipose tissue macrophages may release TNF-α in response to an increase in the availability of free fatty acids (FFAs) [x].

Role of oxidative stress : Oxidative stress develops in NAFLD mostly as a result of increased free fatty acid oxidation by mitochondria, peroxisomes, and microsomes.The imbalance between pro-oxidants, such as RNS (reactive nitrogen species) andROS(reactive oxygen species), and antioxidants in cells constitutes oxidative stress. This imbalance leads to cellular deterioration and, in most cases, cell death [xi].

Insulin resistance and NAFLD: The severity of NAFLD and its etiology appear to be greatly influenced by IR. Cellular insulin signaling may be inhibited by molecular factors(including insulin signaling terminators, IR mediators, and IR-generating factors) that, individually or collectively, change the biological activity of any of the insulin cascade's components, leading to IR and its hepatic component, NAFLD. Free fatty acid excess, ROM overload brought on by oxidative stress, and changes in adipocytokine secretion and activity are IR-inducing variables that start the impairment of insulin signaling implicated in the development of NAFLD [xii]

Significance of cytokines and adipokines: Inflammation, immunity, lactation, hematopoiesis, body growth, adiposity, and other basic biological processes are all mediated by cytokines. Through the promotion of hepatic inflammation, apoptosis, and the creation of fibrosis, cytokines may actively contribute to the onset and probable progression of NAFLD. Cytokines like TNF-α, IL-6, IL-1, and IL-18 are responsible for the pathogenesis of NAFLD [xiii].

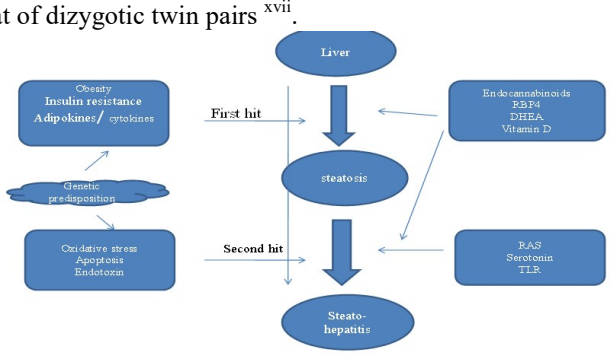
TNF-α: TNF-α is considered an essential cytokine that is directly linked to the development of NAFLD. Kupffer cells (KCs), the resident hepatic cells in NAFLD, produce TNF-α in the liver as well as immune cells that invade the liver because of steatosis. In the case of obesity, dysfunctional visceral adipose tissue plays a crucial role in the creation and release of TNF-α, which is primarily produced by immune cells that infiltrate the adipose tissue. Together with other cytokines and adipokines, TNF-α of extra-hepatic origin is transported to the liver through the systemic circulation and may also have an impact on the onset and severity of NAFLD [xiv].

***Adiponectin*:** Adipocytes are the primary source of the cytokine adiponectin**.** It is a "protective" adipocytokine as it controlsthe metabolism of lipids and glucose and inflammation as well by preventing the production ofNF-κBand TNF-α in macrophages. Insulin resistance and adiponectin levels are inversely correlated, and adiponectin levels are lower in obese individuals [xv].Adiponectin levels rise in NASH patients receiving thiazolidinedione therapy, indicating improvement in hepatic steatosis, necroinflammation, and—most importantly—fibrosis.

Endocannabinoids in Nonalcoholic Fatty Liver Disease and NonalcoholicSteatohepatitis: The endocannabinoidshave a variety ofactions like those of marijuana, and are mediated by CB1 and CB2 receptors.NAFLD and liver fibrosis linked to chronic liver injury are promoted by endogenous activation of CB1(cannabinoids) receptors. Endogenous endocannabinoids and the CB2 receptor are both involved in preventing liver fibrosis, making them crucial therapeutic targets for this reversible condition.Cirrhosis and end-stage liver disease, which are both irreversible, can eventually result from liver fibrosis [xvi].

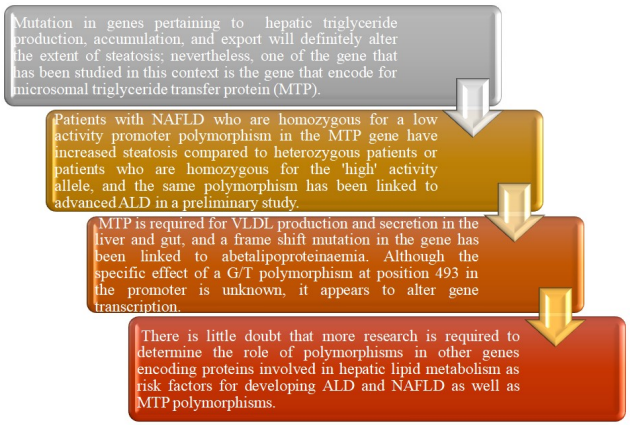
Evidence that NAFLD risk is influenced by genetics: The primary source of evidence for a genetic component of ALD is twin research that found that monozygotic twin pairs had a

concordance risk for alcoholic cirrhosis that was thrice than that of dizygotic twin pairs [xvii].



The different concordance rates for alcoholism specifically did not fully account for this variance in concordance rates. The significance of genetic variables in NAFLD was proposed by two recent family clustering studies. Struben *et al*. discovered a relationship between NASH and cryptogenic cirrhosis in seven of eight cases investigated [xviii], whereas Willner *et al*. discovered that 18% of 90 patients with NASH had an afflicted first-degree relative [xix]. Undoubtedly, this grouping might simply reflect the well-established heredity of the risk factors for NAFLD—obesityand insulin resistance [xx].

Genes that affect the degree of steatosis: The likelihood of nonalcoholic fatty liver disease (NAFLD) may be influenced by factors impacting the severity of steatosis, considering its significance in the etiology of progressive liver disease [xxi]. This category clearly includes genetic and environmental variables that influence the degree of obesity, as well as functional polymorphisms in genes encoding enzymes involved in hepatic lipid metabolism.Gene polymorphisms pertaining to hepatic triglyceride production and accumulation will affect the severity of steatosis and, given the potential significance of steatosis in the etiology of advanced illness, the risk of NASH and steatofibrosis [xxii].



Genetic factors that affect fatty acid oxidation: Adequate FA oxidation is necessary to avoid fat buildup in the liver.Therefore, increased fatty acid oxidationis responsible for the development of oxidative stress [xxiii, xxiv] It is clear that mitochondrial fat oxidation is necessary for the development of inflammation and fibrosis since children with hereditary abnormalities in mitochondrial beta-oxidation exhibit steatosis but not NASH. Gain-of-function polymorphisms in the genes encoding the proteins involved in peroxisomal and microsomal fat oxidation, as both have the potential to produce ROS, should be expected to predispose to NASH.It is believed that loss-of-function polymorphisms affecting them may lead to the development of NASH because they helpprevent excessive mitochondrial load during periods of high FFA supply.A research investigation showed animal models devoid of the fatty acyl-CoA oxidase (AOX)[9]gene, the primaryenzyme of the peroxisomal beta-oxidation pathway, exhibit majormicrovesicular NASH support concepts. Similar challenges arise when analyzing an initial study that suggests a variation in the PPAR-αgene is connected to NASH. PPAR-α controls the transcription of numerous genes that code for enzymes that oxidize fatty acids in the mitochondria, peroxisomes, and microsomes.Functional information on the mutation is now somewhat inconsistent. PPAR-α activation by adiponectin and its protective effects against steatosis, as well as studies in PPAR-α knockout mice [xxv], however, imply that anyPPARA alteration connected to NAFLD ought to be followed by either reduced gene expression or loss of function.

Genes affecting fibrosis severity: Clearly, the genes that code for proteins are related to hepatic fibrogenesis and may contribute to the fibrosis associated with NAFLD. The polymorphic genes for transforming growth factor (TGF)-1, PPAR, and variousfibrogenic adipocytokines, such as angiotensin II, would be obvious possibilities. Only one study in this field indicates that "fibrosis" genes may be associated with an increased risk of developing advanced fatty liver disease.Advanced fibrosis may be more likely to develop in obese individuals who have high levels of both TGF-1 and angiotensinogen-producing polymorphisms [xxvi].

Genes that affect the severity of oxidative stress: The HFE genes producing proteins associated with the body's antioxidant defense mechanisms may affect the severity of oxidative stress.HFE was the subject of a preliminary Australian investigation, which revealed that 31% of 51patients with NASH and only 13% of controls had minimum 1 copy of the C282Y HFE mutation [xxvii].The severity of fibrosis and an elevated hepatic iron index (HII) were also linked to the mutation. The second prevalent HFE variant, H63D, was not associated with anything in this investigation [xxviii]. Considering the endogenous antioxidant defense mechanisms, a recent preliminary investigation found a link between a variation in the mitochondrial superoxide dismutase (SOD2) targeting sequence and the degree of fibrosis in NAFLD patients [xxiv].

TNF-α production or effects are influenced by certain genes: Regarding the genes in the last group, those affecting the actions of TNF-α, a promoter polymorphism at position -238 in the TNF-α gene, are being linked to NASH. Contradictory functional data are available for this polymorphism. Gene polymorphisms affecting the function of TNF-αare related to research in NAFLD on the low activity promoter polymorphism in the gene producing the anti-inflammatory cytokine interleukin (IL)-10[1].

GWAS on NAFLD: There have been a number of candidate gene studies and a small number of GWAS (genome-wide association studies) in NAFLD populations as a result of the heritability and complexity of NAFLD [xxx].

PNPLA3 (PAPTATIN-LIKE PHOSPHOLIPASE DOMAIN CONTAININGPROTEIN 3): PNPLA3, the first variant discovered through GWAS, has shown a strong and consistently reproduced connection with NAFLD in numerous studies.

Table 1. Genes associated with NAFLD progression (Rives, 2020)

Gene	Biological effect	Effect on NAFLD
PNPLA3	Metabolism of triglycerides	Increase incidence, progression, and HCC risk
MBOAT7	Phospholipid metabolism	Increase incidence, progression, and HCC risk
TM6SF2	Very Low Density lipoprotein secretion	Increase incidence and progession
Glucokinase regulator (GCKR)	De novo lipogenesis	Increase incidence and progession
17β-hydroxysteroid dehydrogenase 13(HSD17B13)	Possibly lipid - related inflammation	Decrease progression
Interleukin – 28B (IL28B)	Innate immunity	Decrease fibrosis
Superoxide dismutase 2	Oxidative stress protection	Increase fibrosis

The hepatic and adipose tissues exhibit significant expression levels of PNPLA3 [xxxi]. The most reliable genetic variant linked to non-alcoholic fatty liver disease is PNPLA3, commonly known as adiponutrin. In the initial GWAS, a PNPLA3 variation was linked to the amount of fat in the liver [xxxii.] The PNPLA3 variant is specifically linked to increased hepatic fat without having a significant direct effect on adiposity or features of insulin resistance. This variant makes people more vulnerable to the full range of liver damage caused by NAFLD, from fatty liver to inflammation and NASH to fibrosis and cirrhosis [xxxiii].

TM6SF2(Transmembrane 6 Superfamily Member 2): A protein called transmembrane 6 superfamily member 2 (TM6SF2), which is confined to the hepatocytes' Golgi apparatus and ER, is responsible for the elevated production of TGs-rich lipoproteins from hepatocytes through the secretion pathway of very low-density lipoprotein[7]. Recent GWAS studies have demonstrated a relationship between the SNP rs58542926 C>T in the transmembrane 6 superfamily member 2 (TM6SF2) gene and the severity of NAFLD.One variant in TM6SF2 is linked with a reduced level of mRNA of TM6SF2in the liver. Reduced levels were not associated with cell damage or proliferation but correlated with increased hepatocytic lipid droplet size and changed expression of many genes involved in triglyceride synthesis (ACSS2, PNPLA3, DGAT1, and DGAT2) [xxxiv].

GCKR: By regulating the flux of GCK (glucokinase) between the nucleus and cytoplasm, GCKR plays a crucial role in hepatic glucose uptake[xxxv]. When exposed to fructose-6-phosphate, the P446L variant of GCKR expresses a loss-of-function protein that cannot restrict glucokinase. Because of this, the P446L variation of GCKR is linked to higher hepatic glucose absorption, which may then promote de novo lipogenesis and thus result in lower levels of insulin and blood glucose [xxxvi]. It has been described that the interaction of PNPLA3 and GCKR minor alleles can explain up to 30% of the hepatic fat levels in obese children. Both raised serum triglyceride levels and an increased risk of fibrosis in NAFLD patients are linked to the GCKR P446L variant.In patients with non-alcoholic fatty liver disease (NAFLD), GCKR variants are now linked to fibrosis[19].

MBOAT7: Lysophopshoplipid acyltransferase, also known as MBOAT7, catalyzes the Lands cycle's acyl chain remodeling of phosphatidylinositols by attaching arachidonic acid to lysophosphatidylinositol and lowering levels of free arachidonic acid. Arachidonic acid induces hepatocyte apoptosis, which leads to inflammation and fibrosis in the liver. The variant of MBOAT7 is linked to an increase in hepatic fibrosis and inflammation in NAFLD patients.

SOD2 (Superoxide dismutase 2): Superoxide dismutase, an enzyme dependent on manganese in the mitochondria, which is encoded for the superoxide dismutase 2 (SOD2) gene, is a protein that guards cells against damage brought on by superoxide radicals [xxxvii]. It's interesting to note that the development from simple steatosis to steatohepatitis is thought to be influenced by oxidative stress [xxxviii].

CONCLUSION

Genes are undoubtedly important disease modifiers influencing the event and severity of NAFLD (nonalcoholic fatty liver disease), according to the complicated pathogenetic riddle surrounding the condition. Through their interactions with environmental variables, genes are essential for the development and continuation of NAFLD (non alcoholic fatty liver disease). PNPLA3 polymorphisms are now the most reliable susceptibility altering factors for progressive hepatic damage and steatosis. But via GWAS research, a number of other genetic variations that cause steatosis and steatohepatitis have been found, and risk factors for developing NASH have been verified.

REFERENCES

[i]Day C.P. “The potential role of genes in nonalcoholic fatty liver disease” , Clin Liver Dis 8(2004); 673- 691

[ii]Marchesini G, Brizi M, Morselli-Labate AM, *et al*. Association of nonalcoholic fatty liver disease with insulin resistance. Am J Med 1999; 107(5): 450-5.

[iii]Yamaguchi K, Yang L, McCall S, *et al*. Inhibiting triglyceride synthesis improves hepatic steatosis but exacerbates liver damage and fibrosis in obese mice with nonalcoholic steatohepatitis. Hepatology 2007; 45(6): 1366-74.

[iv]Mashek DG, Khan SA, Sathyanarayan A, Ploeger JM, Franklin MP. Hepatic lipid droplet biology: Getting to the root of fatty liver. Hepatology 2015; 62(3): 964-7.

[v]Alba, L. M., &Lindor, K. (2003). Non-alcoholic fatty liver disease. *Alimentary pharmacology & therapeutics*, *17*(8), 977-986.

[vi]Erickson, S. K. (2009). Nonalcoholic fatty liver disease. *Journal of lipid research*, *50*, S412-S416.

[vii]Sookoian, S., & Pirola, C. J. (2017). Genetic predisposition in nonalcoholic fatty liver disease. *Clinical and molecular hepatology*, *23*(1), 1.

[viii]Arab, J. P., Arrese, M., & Trauner, M. (2018). Recent insights into the pathogenesis of nonalcoholic fatty liver disease. *Annual Review of Pathology: Mechanisms of Disease*, *13*, 321-350.

[xi]Rives, C., Fougerat, A., Ellero-Simatos, S., Loiseau, N., Guillou, H., Gamet-Payrastre, L., & Wahli, W. (2020). Oxidative stress in NAFLD: role of nutrients and food contaminants. *Biomolecules*, *10*(12), 1702

[x]de Alwis, N. M. W., & Day, C. P. (2007, August). Genetics of alcoholic liver disease and nonalcoholic fatty liver disease. In *Seminars in liver disease* (Vol. 27, No. 01, pp. 044-054).

[xi]Hrubec, Z., & Omenn, G. S. (1981). Evidence of genetic predisposition to alcoholic cirrhosis and psychosis: twin concordances for alcoholism and its biological end points by zygosity among male veterans. *Alcoholism: Clinical and Experimental Research*, *5*(2), 207-215

[xii]Polyzos, S. A., Kountouras, J., & Zavos, C. (2009). Nonalcoholic fatty liver disease: the pathogenetic roles of insulin resistance and adipocytokines. *Current molecular medicine*, *9*(3), 299-314.

[xiii]Braunersreuther, V., Viviani, G. L., Mach, F., & Montecucco, F. (2012). Role of cytokines and chemokines in non-alcoholic fatty liver disease. *World journal of gastroenterology: WJG*, *18*(8), 727.

[xiv]Vachliotis, I. D., & Polyzos, S. A. (2023). The role of tumor necrosis factor-alpha in the pathogenesis and treatment of nonalcoholic fatty liver disease. *Current Obesity Reports*, 1-16.

[xv]Freitas Lima, L.C.; Braga, V.A.; do Socorro de Franca Silva, M.; Cruz, J.C.; Sousa Santos, S.H.; de Oliveira Monteiro, M.M.; Balarini, C.M. Adipokines, diabetes and atherosclerosis: An inflammatory association. *Front. Physiol.* 2015, *6*, 304.

[xvi]Basu, P. P., Aloysius, M. M., Shah, N. J., & Brown Jr, R. S. (2014). The endocannabinoid system in liver disease, a potential therapeutic target. *Alimentary pharmacology & therapeutics*, *39*(8), 790-801.

[xvii]Struben VMD, Hespenheide EE, Caldwell SH. Nonalcoholic steatohepatitis and cryptogenic cirrhosis within kindreds. Am J Med 2000;108:9–13

[xviii]Petta, S., Muratore, C., & Craxì, A. (2009). Non-alcoholic fatty liver disease pathogenesis: the present and the future. *Digestive and Liver Disease*, *41*(9), 615-625.

[xix]Willner IR, Waters B, Patil SR, *et al*. Ninety patients with nonalcoholic steatohepatitis: insulin resistance, familial tendency, and severity of disease. Am J Gastroenterol 2001;96:2957–2961

[xx]De Alwis, N.M.W.,& Day, C.P.(2007,August) " Genetics of alcoholic and non alcoholic liver disease", Seminars in liver disease, volume 27;44-54.

[xxi]Day CP, James OF. Hepatic steatosis: innocent bystander or guilty party? Hepatology 1998;27:1463–1466

[xxii]Day C.P. "Genes or environment to determine alcoholic liver disease and non – alcoholic fatty liver disease", Retrieved from review article.

[xxiii]Miele L, Grieco A, Armuzzi A, Candelli M, Forgione A, Gasbarrini A, *et al*. Hepatic mitochondrial beta-oxidation in patients with nonalcoholic steatohepatitis assessed by 13C-octanoate breath test. Am J Gastroenterol 2003;98:2235 – 6.

[xxiv]Day CP. Pathogenesis of steatohepatitis. Ballieres Best Pract Res Clin Gastroenterol 2002;16: 663 – 78

[xxv]Ip E, Farrell G, Robertson G, *et al*. Central role of PPARdependent hepatic lipid turnover in dietary steatohepatitis in mice. Hepatology 2003;38:123–132

[xxvi]Fan CY, Pan J, Usuda N, Yeldandi AV, Rao MS, Reddy JK. Steatohepatitis, spontaneous peroxisome proliferation and liver tumors in mice lacking peroxisomal fatty acyl-CoA oxidase. Implications for peroxisome proliferator-activated receptor alpha natural ligand metabolism. J Biol Chem 1998;273:15639 – 45

[xxvii]Naik Adviti, Genomic aspects of NAFLD pathogenesis ; Volume 102,Issue 2,August 2013; 84 – 95

[xxviii]George DK, Goldwurm S, Macdonald GA, Cowley LL, Walker NI, Ward PJ, *et al*. Increased hepatic iron concentration in nonalcoholic steatohepatitis is associated with increased fibrosis. Gastroenterology 1998;114:311 – 8

[xxix]Bonkovsky HL, Jawaid Q, Tortorelli K, LeClair P, Cobb J, Lambrecht RW, *et al*. Nonalcoholic steatohepatitis and iron: increased prevalence of mutations of the HFE gene in nonalcoholic steatohepatitis. J Hepatol 1999;31:421 – 9.

[xxx]Saksena S, Daly AK, Leathart JB, Day CP. Manganese dependent superoxide dismutase (SOD2) targeting sequence polymorphism is associated with advanced fibrosis in patients with non alcoholic fatty liver disease. J Hepatol 2003;38(Suppl 2):47

[xxxi]Kim DY, Park JY. Genetic risk factors associated with NAFLD. Hepatoma Res 2020;6:85. http://dx.doi.org/10.20517/2394-5079.2020.96

[xxxii]Seko, Y., Yamaguchi, K. & Itoh, Y. " The genetic backgrounds in nonalcoholic fatty liver disease". *Clin J Gastroenterol* 11, 97–102 (2018) . https://doi.org/10.1007/s12328-018-0841-9

[xxxiii]Valenti, L. V., & Baselli, G. A. (2018). Genetics of nonalcoholic fatty liver disease: a 2018 update. *Current pharmaceutical design*, *24*(38), 4566-4573.

[xxxiv]Severson, T. J., Besur, S., & Bonkovsky, H. L. (2016). Genetic factors that affect nonalcoholic fatty liver disease: A systematic clinical review. *World journal of gastroenterology*, *22*(29), 6742

[xxxv]Raimondo A, Rees MG, Gloyn AL. Glucokinase regulatory protein: complexity at the crossroads of triglyceride and glucose metabolism[J]. Curr Opin Lipidol, 2015, 26(2): 88–95

[xxxvi]Beer NL, Tribble ND, McCulloch LJ, *et al*. The P446L variant in GCKR associated with fasting plasma glucose and triglyceride levels exerts its effect through increased glucokinase activity in liver[J]. Hum Mol Genet, 2009, 18(21): 4081–4088

[xxxvii] Danford, C. J., Yao, Z., & Jiang, Z. G. (2018). Non-alcoholic fatty liver disease: a narrative review of genetics. *Journal of biomedical research*, *32*(6), 389.

[xxxviii]Anstee QM, Seth D, Day CP, Genetic Factors That Affect Risk of Alcoholic and Non-Alcoholic Fatty Liver Disease, Gastroenterology (2016), doi: 10.1053/j.gastro.2016.01.037.

****