Boffin Access
FOSTERING KNOWLEDGE

Open Access Journal | Volume: 3.3

International Journal of Clinical and Medical Cases

ISSN 2517-7346

An Update on Further Progression of NAFLD, NASH with Prospective Therapies Like L-Carnitine (LC), Nicotinamide Ribose (NR) Combination, as well as Apical Sodium Dependent Bile Acids Transporter (ASBT) or Volixibat and Silybin as Alternatives

Kulvinder Kochar Kaur[1*]
Gautam Allahbadia[2]
Mandeep Singh[3]

[1]Scientific Director of Human Reproduction, Punjab, India
[2]Scientific Director, Ex-Rotunda-A Centre for Human Reproduction 672,Kalpak Garden, Perry Cross Road, Mumbai, India
[3]Consultant Neurologist, Swami Satyanand Hospital, Near NawiKachehri, Baradri, Ladowali Road,Jalandhar, Punjab, India

Introduction

Overweight and obesity in adults and children has escalatedat an astonishing rate in the past few decades bringing in a lot of the cost burden [1]. Increased Adipose Tissue (AT) as well as ectopic fat collection is typical properties of obesity which results in associated comorbidities like, Type 2 Diabetes Mellitus (T2DM), Hypertension, dyslipidemia.Recently Nonalcoholic Fatty Liver Diseases (NAFLD) has become the commonest chronic liver disease correlated with obesity [2]. Right now the therapy of obesity concentrates on lifestyle changes which include exercise.No reduction in obesity incidence has occurred in spite of lifestyle as well as diet changes over the past 3 decades [1]. We have reviewed multiple ways of treating obesity medically that includes Qsymia,Contrave, Thylakoids,GLP-1 agonists ,BAT thermogenesis enhancement utilizing mirabegron, probiotics etc but none have been of help in maintenance of weight reduction for long term other than Bariatric Surgery(BS) [3-9]. Further with escalating obesity liver disorders are increasing remarkably with no effective way of treating them .All over the world work is being done at war footing with the enhanced rates of liver transplants needed inthose under 50 yrs [10,11]. Thus we have further tried to present the advances done in working out some medical therapy to prevent propagation of NAFLD and halt it further to not progress till Nonalcoholic Steato Hepatitis (NASH) level,cirrhosis and Hepatocellular Carcinoma (HCC).

Methods

Thus we did an update on the further advances that are being made in treating NAFLD, NASH and carried out a PubMed search using the MeSH Terms NAFLD; NASH; HCC; Preventive or curative therapies.

Results and Discussion

We came across 377 articles out of which we chose 70 articles for updating the earlier information we had reviewed.No meta-analysis was done.

Role of carnitine shuttle and targeting lipid peroxidation

L-Carnitine(LC) has a great part in oxidative metabolism as it is needed for moving Long Chain Fatty Acids (LCFA's) from the cytoplasm into the mitochondrial matrixin which β-oxidation takes place. For getting shifted these LCFA require activation into acyl CoA's and get converted into acyl carnitines for movement across mitochondrial membrane. These acyl carnitines are under C22 in length can get into mitochondrial matrix [12], where get exchanged with free carnitine, they get converted back to acyl CoA's and thus can get utilized for β-oxidation [13]. Alsocarnitine is needed for shifting the end products of peroxisomal β-oxidation,medium as well asshort chain acyl CoA's,out of the peroxisomes for promotingmoremitochondrial processing [14]. Decrease in carnitine amounts correlates with Insulin Resistance (IR) as well as Diet Induced Obesity (DIO) and was pointed to be secondary to prolonged lipid excess, impaired energy metabolism,associated with fat oxidation that is not complete [15].On the other hand, LC administration in obese rats was demonstrated to reinstate carnitine amounts and thereby enhancing metabolic working [15] (Figure1).

Nicotinamide Ribose (NR) is a molecule present within diet that is supposed to be the molecule that is responsible for synthesis of nicotinamide adenine nucleotide (NAD+) which increases oxidative metabolism and in mice has been demonstrated to protect HFD induced obesity within mice [16]. Reducing equivalents are supplied by NAD+ for oxidative phosphorylation,that is necessary for oxidative metabolism as well as metabolic

Article Information

DOI: 10.31021/ijcmc.20203138
Article Type: Case Report
Journal Type: Open Access
Volume: 3 Issue: 3
Manuscript ID: IJCMC-3-138
Publisher: Boffin Access Limited

Received Date: 06 January 2020
Accepted Date: 27 January 2020
Published Date: 29 January 2020

***Corresponding author:**

Kulvinder Kochar Kaur
Scientific Director of Human Reproduction
721, G.T.B. Nagar, Jalandhar
Punjab, India
Fax: 91-181-4613422
Tel: 91-181-9501358180
E-mail: kulvinder.dr@gmail.com

Citation: Kochar KK, Allahbadia G, Singh M. An Update on Further Progression of NAFLD, NASH with Prospective Therapies Like L-Carnitine (LC), Nicotinamide Ribose (NR) Combination, as well as Apical Sodium Dependent Bile Acids Transporter (ASBT) or Volixibat and Silybin as Alternatives. Int J Clin Med Cases. 2020 Jan;3(3):138

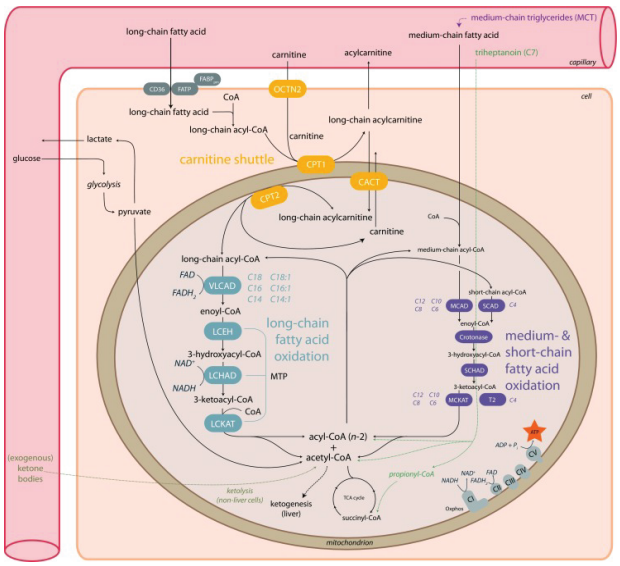

Figure 1: Courtesy ref number16 -Schematic representation of mitochondrial fatty acid oxidation in humans

Description: Long-chain fatty acids enter the cell from the bloodstream and enter the mitochondria through the carnitine shuttle, followed by a step-wise degradation involving a series of enzymes of the long-chain fatty acid oxidation machinery resulting in the production of acetyl-CoA. Potential treatments to produce acetyl-CoA independent of the fatty acid oxidation enzymes are indicated. These include medium-chain triglycerides, ketone bodies and triheptanoin. Abbreviations: CI-V, Complex I-V; CACT, Carnitine Acylcarnitine Translocase; CD36, Cluster of Differentiation 36; CoA, Coenzyme A; CPT1, Carnitine Palmitoyl Transferase type 1; CPT2, Carnitine Palmitoyl Transferase type 2; FABP pm, plasma membrane-associated Fatty Acid Binding Protein; FATP, Fatty Acid Transport Protein; LCEH, Long-Chain Enoyl-CoA Hydratase; LCHAD, Long-Chain 3-Hydroxyacyl-CoA-Dehydrogenase; LCKAT, Long-Chain Ketoacyl-CoA Thiolase; MCAD, Medium-Chain Acyl-CoA Dehydrogenase; MCKAT, Medium-Chain 3-Ketoacyl-CoA Thiolase; OCTN2, Organic Cation Transporter 2; SCAD, Short-Chain Acyl-CoA Dehydrogenase; SCHAD, Short-Chain 3-hydroxyacyl-CoA Dehydrogenase; T2, acetoacetyl-CoA thiolase.

homeostasis .In obesity along with other components of Metabolic Syndrome (MetS) like Type 2 Diabetes Mellitus (T2DM) as well as NAFLD [17,18], pointing that with external supplementation to escalate NAD+ levels might help in reducing these conditions.Further NAD+ helps in avoiding extent of injury secondary to Oxidative Stress (OS) [17]. In Obesity as well as NAFLD formation role of Oxidative Stress (OS) has been documented [19]. An escalation of 4-hydroxy nonenal (4-HNE), that represents a marker of Oxidative Stress (OS)-stimulated lipid peroxidation [20] which takes place in NAFLD subjects [21,22]. Thus Salic et al. analyzed if giving a combination of l-Carnitine (LC) and Nicotinamide Ribose (NR), both parts of natural substances which might increase transfer of FA'sinside mitochondrial membrane and thus enhance NAD+) amounts, that are known to be essential for β-oxidation as well as tricarboxylic acid cycle(TCA) respectively (Figure 2). Ldlr-/-Leiden micegot treated With High Fat Diet (HFD) that had administration of (LC;0.4%w/w), (NR;0.3%w/w) or both (COMBI) for 21 weeks. Levels of LC that were decreased by HFD got to normal after LC administration. Similarly with NR administration increased its plasma amounts of metabolism products suggesting proper administration. Inspite of food orally along with locomotion were relatively same in all groups, COMBItherapy markedly ameliorated HFD stimulated enhancement of body weight,fat mass increase which was -17% as well as hepatic steasosis that reduced by 22%. Further, both NR and COMBI decreased liver 4-HNE adducts. On checking the upstream regulator

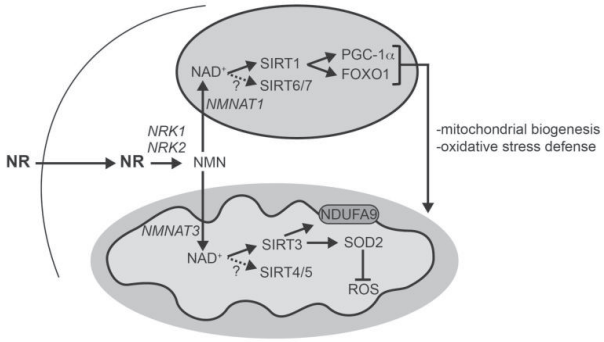

Figure 2: Courtesy ref no-13Schematic representation of the different actions of NR in metabolic homeostasis

Description: The scheme summarizes the hypothesis by which NR supplementation would increase NAD^+ content in key metabolic tissues, leading to SIRT1 and SIRT3 activation and the deacetylation and modulation of the activity of key metabolic regulators. This model does not rule out the participation of additional mechanisms of action for NR to achieve its beneficial effects. Abbreviations can be found in the text and enzymes are indicated in italics.

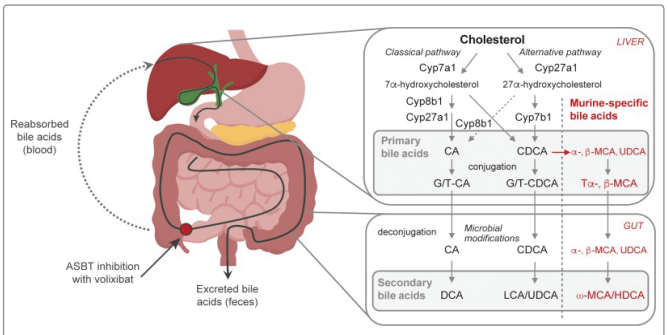

Figure 3: Primary bile acids are synthesized from cholesterol in the liver, via either the classical pathway, which produces CA, or the alternative pathway, which produces CDCA, or, in mice only, α- or β-MCA.Bile acids are conjugated with glycine or taurine (predominantly taurine in mice) before being released into the bile [30]. In the gut, bile acids are deconjugated and metabolized into secondary bile acids (UDCA is considered a secondary bile acid in humans but a primary bile acid in mice). About 95% of bile acids are reabsorbed from the gut and transported back to the liver via the hepatic portal vein, and the remainder is excreted. The hydrophilicity of the common free and conjugated bile salts decreases in the order UDCA > CA > CDCA > DCA) > LCA, and taurine-conjugated > glycine-conjugated > free species. ASBT inhibition with volixibat blocks the reabsorption of bile acids and increases their excretion, stimulating the liver to synthesize more bile acids from cholesterol. ASBT: apical sodium-dependent bile acid transporter, CA: cholic acid, CDCA: chenodeoxycholic acid, Cyp: cytochrome P450 family, DCA: deoxycholic acid, G: glycine, HDCA: hyodeoxycholic acid, LCA: lithocholic acid, MCA: muricholic acid, T: taurine, UDCA: ursodeoxycholic acid

gene analysis showed that COMBI finished the negative actions of HFD on liver metabolism pathways correlated controllers like ACOX, SCAP, SREBP, PPARGC1B and INSR. Thus a combination therapy of LC and NR confers protection on metabolic paths and might reflect a prospective therapy for obesity caused by HFD as well as NAFLD [23].

Role of targeting bile acid metabolism

Nonalcoholic Steato Hepatitis (NASH) represents a severe and mostly propagating type of NAFLD [24-26] for which no approved

medical therapy exists pointing to severe need for developing one [27]. On histology, hepatic steatosis, lobular inflammation with damage of the liver cells injury or ballooning that causes cell necrosis either with or without fibrosis [28]. Since classically NASH remains silent a liver biopsy is needed to pick it up early [28,29]. It might cause cirrhosis, Hepatocellular Carcinoma (HCC), [30] as well as liver failure [31,32], being the biggest cause for liver transplantation in US adults below 50years [33]. There is complicated pathophysiology of NASH along with multifactorial [reviewed earlier by us unpublished, under review] having multiple factors in combination like genetic, environmental, Gut Microbiota (GM),. Additionally dysfunctional metabolism of bile acids, lipids that include cholesterol as well as Insulin Resistance (IR) [34]. Excessive synthesis as well as decreased excretion of cholesterol from liver ends up in the collection of free cholesterol [35] that correlates with hepatic injury, mainly via interfering of mitochondrial function [36] as well as facilitation of Oxidative Stress (OS) [37].

Escalated serum insulin as well as triglycerides associated with increased amounts of Very Low Density Lipoproteins (VLDL) cholesterol and decreased High Density Lipoprotein (HDL) cholesterol [37,38]. In NASH subjects IR as well as hyperlipidemia is frequent just like visceral obesity accompanied with MetS [24]. White Adipose Tissue (WAT) enhancement might aid in disease propagation via synthesis of adipokines, inflammatory cytokines as well as lipids [39].

Commonly serum bile acids are increased in NASHsubjects, with levels 2 times >than normal [40]. With bile acids amounts elevated inflammatory responses might present with Oxidative Stress (OS),cell death paths that might lead to liver injury [41,42]. Primary bile acids get manufactured via cholesterol in the liver and shifted to gut via bile where they might be metabolized to secondary bile acids by bacteria (Figure 3) [38,43]. Secondary bile acids like Lithocholic Acid (LCA) and Deoxy Cholic Acid (DCA), have > hydrophobicity [42,43] and might stimulate inflammation, enhance Reactive Oxygen Species (ROS),and stimulate necrotic pathways [42-44].

Although the rates are constantly escalating we don't have any effective therapy but for diet changes and exercise [25,45]. Lot of pharmacological therapies that hold a promise is being studied [45]. One strategy is to block the enterohepatic recirculation of bile acids back to liver by blocking the Apical Sodium Dependent Bile Acids Transporter (ASBT) [46]. The ASBT is a transmembrane protein present in the luminal surface of ileal enterocytes.Roughly 95% of the bile acids pool gets resorped in the terminal ileumand then shifted back to liver, and the rest gets excreted via feces [43,47] (Figure 3).

Blocking bile acids recycling via inhibition of ASBT enhances the fecal excretion [46,48] and is believed to induce the liver for more bile acids manufacture from hepatic cholesterol for sustaining the bile acids homeostasis [49].In mice inhibition of ASBT escalated hepatic expression of cholesterol 7α hydroxylase (Cyp 7a1) [49], a rate limiting enzyme in bile acids manufacture [50,51]. Volixibat Potassium (SHP26,earlier called LUM002 now labeled Volixibat), a minimally absorbed ASBT inhibitor [46,52] was partly used for a phase 2 clinical study in patients with NASH (ClinicalTrial.gov) Identifier NCT02787304 in parallel with the below described mouse study by Salic et al. [53].

Hence Salic etal. tried to test the probable disease ameliorating action of ASBT inhibitor volixibat (5,15 and 30mg /kg) in HFD fed Ldlr-/-Leiden mice over 24 weeks .Plasma as well as fecal bile acids amounts, plasma insulin ,lipids , liver enzymes were evaluated. Ultimate examination was liver histology, intra hepatic lipids, mesenteric WAT mass as well as liver gene profiles testing. Volixibat escalated the total amount of fecal bile acids. At the maximum dose , Volixibat significantly ameliorated the HFD stimulated increased hepatocyte hypertrophy, liver triglycerides as well ascholesteryl ester levels as well as mesenteric WAT getting accumulated. Nonalcoholic fatty liver diseases scores (NAS) were significantly low in Volixibat treated mice act HFD controls. Gene profiling demonstrated that Volixibat finished the inhibitory action of HFD on metabolic controllers like peroxisome proliferator activated peroxisome proliferator activated receptor – γ coactivators (PGC1s), insulin receptor as well as sterol regulatory element binding transcription factor 2. Thus Volixibat might be effective physiologically as well as regarding metabolic part of NASH pathophysiology [53].

Role of Silybin

Globally the incidence of metabolic disorders like Nonalcoholic Fatty Liver Diseases (NAFLD), Type 2 Diabetes Mellitus (T2DM), as well as obesity has presented in anepidemic fashion worldwide. NAFLD incidence is 6-35% (median 20%) worldwide with this number escalating worldover, in view of the diet andlifestylepatterns [54]. This NAFLD prevalence is markedly associated with obesity, T2DM as well as dyslipidemia [55]. The ''multiple hit hypothesis'' is thought to be responsible for the etiopathogenesis of NAFLD, that includes IR, Adipose Tissue(AT) liberated hormones, nutritional factors, GM as well as genetic and epigenetic factors [56,57]. As NAFLD progresses simple steatosis might shift to the more severe form labeled Nonalcoholic Steatohepatitis (NASH) that includes inflammation as well as apoptosis, with or without fibrosis as well as cirrhosis. No proper way of avoiding the occurrence and formation of NASH exists other than diet control.Discovery of new drugs for curing NAFLD and NASH will be very important.

For treating these chronic problems Traditional Chinese Medicine (TCM) have been utilized for long in Chinaas well assome Asian countries for centuries. As no medical therapy from West has been shown to be safe and efficacious for treating NAFLD, Traditional Chinese Medicine (TCM) pose a particular superiority.Silybin is extracted from milk thistle (Sylibummarianum) seeds and is used as a hepato protective agent broadly [58]. Different potential biological actions of Silybin have been looked for, that includes cancer therapy, DM treatment and liver diseases therapy [59,60]. Mode of action includes control of lipid metabolism,antioxidant radical scavenger. Cellular membrane getting stabilized and promotion of ribosomal and RNA synthesis [61,62] has also been documented that silybin manipulated variety of metabolism pathways in acute liver injury [63]. But no metabolism information of Silybin has been totally given that might give us understanding of its mechanism of action.

Metabolomics gives information in a comprehensive manner that regarding endogenous molecules in the body and is very useful in diagnosing the variety of diseases for decreasing the chances of these conditions [64,65]. Moreover biomarkers are used in an unbiased way to for classification of the stage of disease propagation [65,66]. It also lends biomarkers to check the effectiveness of drugs, also labeled as pharmacometabolomics [67,68]. Serum biomarkers, like total cholesterol, triglycerides. C peptide and Glucose Tolerance Test (GTT), have been utilized in diagnosing NAFLD for several years. Right now there are more upcoming biomarkers giving important complementary knowledge, like Apo lipoproteins A1, apolipoprotein B leptin, adiponectin, Free Fatty Acids (FFA), ghrelin and Tumor Necrosis Factor Alpha (TNFα) [69]. But a comprehensive and a total view of metabolites that are altered in NAFLD and following Silybin therapy is not available. Hence Sun et al. [70] tried to evaluate the metabolic control by Silybin of NAFL D.C57BL/6J mice that were given HFD/High cholesterol diet for 8wks and treated with Silybin (50 or 100mg/kg/day) as well as sodium taurourso deoxycholate (TUDCA, 50mg/kg/day) by gavage for the last 4 weeks. Blood biochemical indexes and hepatic lipid measurement along with Oil red O staining of the liver were carried out for analyzing the model as well as the lipid decreasing effect of Silybin and TUDCA. Moreover serum and liver samples were checked utilizing a metabolomics platform on the basis of gas chromatography-mass spectrometry (GC/MS). Multivariate/univariate data analysis and pathway evaluation were utilized for evaluation of different metabolites as well as metabolic pathways. As per the results the mouse NAFLD model got established successfully and Silybin and TUDCA decreased both serum as well as hepatic lipid collection at significant levels. Metabolomics evaluation of serum and liver demonstrated that HFD/High cholesterol diet led

to the anomalous metabolism of metabolites participating in lipid metabolism, polyol metabolism, amino acid metabolism, the urea cycle and the TCA cycle. Silybin and TUDCA therapy together reversed metabolic disorders produced by HFD feeding. Thus concluding that a HFD/High cholesterol diet resulted in anomalous metabolism in the serum and liver of mice, and Silybin therapy improved hepatic lipid collection and manipulated global metabolic pathways that gave a probable way for the multiple target modes of action [70].

Conclusions

Thus, here we have summed up the role of utilizing LC and NR for affecting lipid metabolism by enhancing LC levels and with use of NR how OS is reduced along with reduction of 4-hydroxy nonenal (4-HNE) levels following increase of nicotine adenine diamide on supplementing LC and NR together that might be one strategy for reducing propagation of NAFLD. The further role of Apical Sodium Dependent Bile Acids Transporter (ASBT) inhibitor volixibat by affecting bile acid metabolism that is undergoing phase 2 trials in humans for NASH and further the role of Silybin a product that has been and rosmarinic acid we have researched from TCM. Already probiotics, work on Vitamin D Metabolism, allylisothiocyanate reviewed along with role of the gut microbiota in aetiopathogenesis etc. Hopefully we will find some permanent method for prevention of NAFLD, NASH propagating to cirrhosis HCC, liver failure and avoid need of liver transplantation with incidence increasing with epidemic of obesity and Diabesity and diabetes itself being a causative factor in NAFLD as well as NASH independent of obesity.

References

1. Ng M, FleminT, Robinson M, Thomson B, Graetz N, etal. Global , regional and national prevalenceof overweight and obesity in children and adult during 1980-2013: A Systematicanalysis for the Global Burden of Disease Study 2013. Lancet 2014 Aug;6736:1-16.
2. Younossi ZM. Nonalcoholic fatty liverdisease-A global public health perspective. J Hepatol. 2019 Mar;70(3):531-544.
3. KulvinderKocharKaur, Allahbadia GN, Singh M. An Update on Aetiopathogenesis and Management of Obesity. Obesity and Control Therapies: Open Access. 2016;3(1):1-17.
4. KulvinderKocharKaur, Allahbadia GN, Singh M. Hypothalamic inflammation and glioses as aetiopathogenetic factor in high fat diet induced obesity and variousthera peutic options to resolve it. Obes Res Open J. 2017; 4(2):44-60.
5. Kaur KK, Allahbadia GN, Singh M. Current advances in pathogenesis in obesity: Role of Hypothaalamicglioses. J Obes Weight Loss. 2018 Jan;3:008:1-11.
6. Kaur KK, Allahbadia GN, Singh M. Have Probiotics and Synbiotics passed the test of time to be implemented in management of obesity and related metabolic disorders-a comprehensive review. AdvObes Weight Manag Control. 2019;9(1):21-28.
7. Kaur KK, Allahbadia GN, Singh M. Weight loss Associated with high protein Intake in Obesity:Interactions of Gut Microbiota in Protein Sourcesinfluencing this positive effect. Acta Scientific Nutritional Health 2018 June;2(7):80-89.
8. Kaur KK, Allahbadia GN, Singh M. Rosmarinic Acid-A New Hope for Liver Diseases Like Cirrhosis, Hepatocellular Carcinoma-Needs Translation to Humans. EC Endocrinology and Metabolic Research. 2019 Aug;289-301.
9. Kaur KK, Allahbadia GN, Singh M. Advances in Targeting the Gut Microbiota for Obesity Therapy. Acta Scientific Microbiology. 2019 August;2(9):78-79.
10. Fildes A, Charlton J, Rudisill C, Littlejohns P, Prevost AT, et al. Probability of an obese person attaining normal body weight:Cohort study using electronic health records. Am J Public Health 2015 Sep;105(9)e54-49.
11. Lee Y, Kwon EY, Choi MS. Dietary isoloquiritigenin at a low dose ameliorates insulinresistance and NAFLD in diet induced obesity in C57BL/6J mice. Int J MolSci 2018 Oct;19(10):3281.
12. Schrader M, Costello J, Godinho LF, Islinger M. Peroxisome – mitochondria interplay and disease. J Inherit Metab Dis 2015 Jul;38(4):681-702.
13. Wijburg FA, Wust RCI, Visser G, Wanders RJA, Knottnerus SJG, etal. Disorders of mitochondrial long chain fatty acid oxidation and the carnitine shuttle. Rev Endocr Metab Disord. 2018 Mar;19(1):93-106.
14. Antonenkov VD, Hiltunen JK. Transfer of metabolites across the peroxisomalmembrane. Biochem Biophys Acta. Sep;1822(9):1374-1386.
15. Noland RC, Koves TR, Seiler SE, Lum H, Lust RM, etal. Carnitine insufficiency caused by aging and overnutrition compromises mitochondrial performance and metabolic control. J Biol Chem. 2009 Aug;284(34):22840-22852.
16. Canto C, Houtkooper RH, Pirinen E, Youn DY, Oosterveer MH, etal. The NAD+ precursorNicotinamideriboside enhances oxidative metabolism and protects against high fat diet induced obesity. Cell Metab. 2012;6(15):838-847.
17. Garten A, Schuster S, Penke M, Gorski T, DeGiorgis T, et al. Physiological and pathophysiologicalroles of NAMPT and NAD metabolism. Nat Rev Endocrinol 2015 Sep;11(9):535-546.
18. Yoshino J, Baur JA, Imai SI. NAD+ Intermediates. The biology and therapeutic potential of NMN and NR. Cell Metab. 2018 Mar;27(3):513-528.
19. Bournat JC, Brown CW. Mitochondrial dysfunction in obesity. CurrOpinEndocrinol DiabetesObes. 2010 Oct;17(5):446-452.
20. Zhong H, Yin H. Role of lipid peroxidation derived 4-hydroxy nonenal (4-HNE)in cancer. Focusing on Mitochondria. Redox Biol. 2015; 4:193-199.
21. Seki S, Kitada T, Yamada T, Sakaguchi H, Nakatani K, et al. In situ detection of lipid peroxidation and oxidative DNA damage in Nonalcoholic fatty liver diseases. J Hepatol. 2002 Jul;37(1):56-62.
22. Ore A, Akinloye OA. Oxidative stress and Antioxidant Biomarkers in Clinical and Experimental models of Nonalcoholic fatty liver diseases. Medicina. 2019 Jan;55:(2).
23. Salic K, Gart E, Seidel F, Verschuren L, Caspers M, etal. Combined treatment with L-Carnitine and Nicotinamideriboside Improves Hepatic Metabolism and Attenuates Obesity and Liver Steatosis. Int J Mol Sci. 2019 Sep;20(18):4359.
24. Younossi ZM, Anstee QM, Marietti M, Hardy T, Henry L. Global burden of NAFLD and NASH:trends , predictions , risk factorsand prevention. Nat Rev Gastroenterol Hepatol. 2018Jan;15(1):11-20.
25. Chalassani N, Younossi ZM, Lavine JE, Charlton M, Cusi K, etal. The diagnosis and management ofNonalcoholic fatty liverdiseases:practice guidancefrom the American Association for the Studyof Liver Diseases. Hepatol. 2018 Jan;67(1):328-357.
26. Milic S, Lulic D, Stimac D. Nonalcoholic fatty liver diseases:biochemical, metabolic and clinical presentations. World J Gastroenterol. 2014 Jul;20(28):9330-9337.
27. Borodosi L, Marchignoli F, Petroni ML, Marchesin IG. NASH: A glance at the landscape of pharmacological treatment. Ann Hepatol. 2016 Sep-Oct;15(5):673-681.
28. Chalassani N, Younossi ZM, Lavine JE, Diehl AM, Brunt EM, etal. The diagnosis and management of Nonalcoholic fatty liver diseases: practice guidance from the American Association for the Study of Liver Diseases, and the American GastroenterologicalAssociation. Hepatol. 2012 Jun;55(6):2005-2023.
29. EASL-EASO-EASO Clinical Practice Guidelines for the management of Nonalcoholic fatty liver diseases. J Hepatol. 2016 Jun;64(6):1388-1402.
30. White DL, Kanwal F, El-SerabHB. Association between Nonalcoholic fatty liverdiseases and risk for Hepatocellular cancer, based on systematic review. ClinGastroenterolHepatol.

2012 Dec;10(12):1342-1359.

31. Mc Pherson S, Hardy T, Henderson E, Burt AD, Day CP, et al. Evidence of NAFLD progression from steatosisto fibros ingsteatohepatitis using pairedbiopsies:implications for prognosis and clinical management. J Hepatol. 2015 May;62(5):1148-1155.
32. Wong VW-S, Wong GL-H, Choi-PC-L, Chan AW-H, Li HY, etal. Disease progression of Nonalcoholic fatty liverdiseases: a prospective study of paired liver biopsies at 3 years. Gut. 2010;59(7):969-974.
33. Banini B. Nonalcoholic steatohepatitis(NASH) has surpassed hepatitis C as the leading etiology for listing for liver transplant: implications for NASH in children and young adults. American College of Gastroenterology Annual Scientific Meeting 14-19-october 2016;Las Vegas 2016.
34. Hallibasic E, Claudel T, Trauner M. Bile acid transporters and regulatory nuclear receptors in the liver and beyond. J Hepatol. 2013 Jan;58(1):155-168.
35. Musso G, Gambino R, Cassender M. Cholesterol metabolism and the pathogenesis of Nonalcoholic steatohepatitis. Prog Lipid Res 2013 Jan;58(1):155-168.
36. Mari M, Caballero F, Colell A, Morales A, Caballeria J, etal. Mitochondrial free cholesterol loading sensitizes to TNF and Fas mediated steatohepatitis. Cell Metab. 2006 Sep;4(3):185-188.
37. Siddiqui MS, Fuchs M, Idowu MO, Luketic VA, BoyettS, etal. Severity of Nonalcoholic fatty liverdisease and progression toto cirrhosis are associatedwith atherogeniclipoproteinprofile. Clin Gastroenterol Hepatol. 2015 May;13(5):1000-1008.
38. Arquello G, Balboa E, Arresse M, Zanlugo S. Recent insights on the role of cholesterol in Nonalcoholic fatty liverdisease. BiochimBiophysActa. 2015 Sep;1852(9):1765-1778.
39. Mulder P, Morrison M, Wielinga P, Van Duyvenvoorde W, Koistra T, et al. Surgical removal of inflamed epididymal white adipose tissue attenuates the development of Nonalcoholic steatohepatitis in obesity. Int J Obese(Lond) 2016 Apr;40(4):675-684.
40. Ferslew BX, Xie G, Johnston CK, SuM, Stewart PW, etal. Altered bile acid metabolome in patients with Nonalcoholic steatohepatitis. Dig Dis Sci. 2015 Nov;60(11):3318-3328.
41. Palmer M, Jennings L, Silloerg D, Bliss C, Martin P. et al. A minimally absorbed, oral, apical sodium dependent bile acids transporter (ASBT), increases bile acid excretion, reduces serum lipids, is safe and tolerable in overweight and obese subjects, a population characteristic of NASH. Hepatol. 2016;64(Suppl1):574
42. Perez MJ, Briz O. Bile acid-induced cell injury and protection. World J Gastroenterol. 2009 Apr;15(14):1677-1689.
43. Staels B, Fonseca VA. Bile acid and metabolic regulation: mechanisms and clinical responses to Bile acid sequestration. Diabetes Care. 2009 Nov;32Suppl2:S237-245.
44. Allen K, Jaeschke H, Copple BL. Bile acids induce inflammatory genes in hepatocytes: a novel mechanism of inflammation during obstructive cholestasis. Am J Pathol. 2011 Jan;178(1):175-186.
45. Rotman Y, Sanyal AJ. Current and upcoming pharmacotherapy for Nonalcoholic fatty liver disease. Gut. 2017 Jan;66(1):180-190.
46. Siebers N, PalmerM, Silberg DG, Jennings L, Bliss C, et al. Absorption, distribution, Metabolism and excretion of [14C]-Volixibat in healthy men:phase 1 open label study. Eur J Drug MetabPhsarmacokinet. 2018 Feb;43(1):91-101.
47. Hoffmann AF. The enterohepatic circulation of Bile acids in mammals:forms and functions. Front Biosci(Landmark ed). 2009 Jan;14:2584-2598.
48. Rao A, Kosters A, Mells JE, Zhang W, SetchellKD, etal. Inhibition of ileal bile acid uptake protectsagainst Nonalcoholic fatty liverdisease in high fat fedmice. SciTransl Med. 2016 Sep;8(357):357ra122.
49. Chiang JYL. Bile acid Metabolism andsignaling in liver disease and therapy. Liver Res 2017 Jun;1(1):3-9.
50. Jelinek DF, Andersson S, Slaughter CA, Russell DW. Cloning and regulation of cholesterol 7 alpha – hydroxylase , the rate limiting enzymein Bile acid biosynthesis. J BiolChem 1990 May;265(14):8190-8197.
51. Galman C, Arvidsson I, Angelin B, Rudling M. Monitoringhepatic cholesterol 7 alpha –hydroxylaseactivityby assayof the stable bile acid intermediate7α hydroxyl-4 cholesten -3-one in peripheral blood. J Lipid Res. 2003;44(4):859-866.
52. Tiessen RG, Kennedy CA, Keller BT, Levin N, Acevedo L, etal. Safety , tolerabilityand pharmacodynamics of apicalsodium –dependentbile acid transporter inhibition with Volixibat in healthy adultsand patientswith type 2 diabetes mellitus: a randomized placebo-controlled trial. BMC Gastroenterol. 2018 Jan;18(1):3.
53. Salic K, Kleeman R, Wilkins-Port C, McNutty J, Verschuren L, et al. Apical sodium dependentbile acidstransporter inhibition with Volixibat improves metabolic aspects and components of Nonalcoholic steatohepatitis in Ldlr-/-Leiden mice. PLoS ONE. 2019 June;14(6):e0218459.
54. Bellentani S. The epidemiology of Nonalcoholic fatty liver diseases. Liver Int. 2017 Jan;37(Suppl1):81-84.
55. Samuel VT, Shulman GI. Nonalcoholic fatty liver disease as anexus of metabolic and hepatic diseases. Cell Metab. 2018 Jan;27(1):22-41.
56. Da Silva HE, Teterina A, Comelli EM, Talbi A, Arendt BM, et al. Nonalcoholic fatty liver disease is associated with dysbiosis independent of body mass index and insulin resistance. Sci Rep. 2018 Jan;8(1):1466.
57. Buzzetti E, Pinzani M, Tsochatzis EA. The multiple-hit pathogenesisof Nonalcoholic fatty liverdisease (NAFLD). Metab Clin Exp. 2016 Aug;65(8):1038-1048.
58. Fang YL, Chen H, Wang CL, Liang L. Pathogenesis of Nonalcoholic fatty liverdisease(NAFLD). In children and adolescents from ''two hit theory to multiple-hitmodel. World J Gastroenterol. 2018 Jul;24(27):2974-2983.
59. Federicko A, Dallio M, LoguercioC. Silymarin/Silybin and chronic liver disease: a marriage of many molecules. Molecules 2017 Jan;22(2):191.
60. Kren V, Walterova D. Silybin and Silymarin-new effects and applications. Biomed Papers 2005 Jun;149(1):29-41.
61. Vahabzadeh M, Amiri N, Karimi G. Effects of Silymarin on metabolic syndrome:a review. J Sci Food Agric. 2018 Oct;98(13):4816-4823.
62. Zhang B, XuD, She L, Wang Z, Yang N, et al. Silybin inhibit NLRP3 inflassome assembly through the NAD(+)/SIRT pathway in mice with Nonalcoholic fatty liver disease. FASEB J. 2018 Feb;32(2):757-767.
63. Ni X, Wang H. Silymarin attenuated hepatic steatosis through regulation of lipid metabolism and Oxidative stress in a mouse model of Nonalcoholic fatty liver disease (NAFLD). Am J Transl Res. 2016 Feb;8(2):1073-1081.
64. Liang Q, Wang C, LiB, Zhang AH. Metabolic fingerprinting to understand therapeutic effects and mechanisms of Silybin on acute liver damage in rat. Pharmacogn Mag. 2015 Jul-Sep;11(43):586-593.
65. Wang X, Zhang A, Sun H. Power of Metabolomics in diagnosis and biomarker discovery of hepatocellular carcinoma. Hepatology. 2013;57(5):2072-2077.
66. Zhang A, Sun H, Yan G, Wang P, Han Y, et al. Metabolomics in diagnosis and biomarker discovery ofcolorectal cancer. Cancer Lett. 2014 Apr:345(1):17-20.
67. Shah VO, Townsend RR, Feldman HL, Pappan KL, Kensicki E, et al. Plasma Metabolomic profilesin different stages of CKD. Clin J Am SocNephrol. 2013 Mar;8(3):363-370.
68. Kaddurah-Daouk R, Weinshiliboum RM, Pharmaco Network. Pharmacometabolomics: implications for clinical pharmacology and systems pharmacology. Clin Pharmacol Ther. 2014

Feb;95(2):154-167.

69. Neuman MG, Cohen LB, Neuman RM. Biomarkers in Nonalcoholic fatty liverdiseases. CanJ GastroenterolHepatol. 2014 Dec;28(11):607-618.
70. Sun R, XuD, Wei Q, Zhang B, Aa J, et al. Silybin ameliorates hepatic lipid accumulation and modulates global metabolism in an NAFLD mouse model. Biomedicine and Pharmacotherapy. 2019 Dec;123:109721.