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Advanced Assessment of Hepatic Choline and Lipid Profiling via Magnetic Resonance Spectroscopy among Young Saudi Women: Unraveling Metabolic Dynamics and Implications for Health

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Abstract

Background: Choline, a vital nutrient, and lipids play pivotal roles in liver function and overall metabolic homeostasis. Magnetic resonance spectroscopy (MRS) offers a non-invasive and highly sensitive method for assessing hepatic choline and lipid levels in vivo. This study aims to investigate the correlation between liver choline and lipid levels among young Saudi women.

Methods and Results: This cross-sectional research explored the relationship between liver choline and fat levels, employing MRS to estimate these levels. A total of 29 healthy female students from the radiological sciences department, aged between 18 and 25 years, participated in the study. MRS scans were conducted using a 1.5 tesla Philips scanner using single volume sequence (SVS). The SVS spectroscopy sequence used in this study indicated the presence of lipid peaks within the frequency range of 0.9-1.2 ppm and choline peaks at 3.2 ppm. The Pearson correlation test revealed a statistically significant association between choline and lipid levels in the liver (r=0.367, *P*=0.025).

Conclusion: Investigating the relationship between liver lipids and choline levels is crucial for understanding liver health, identifying metabolic dysregulation, informing diagnostic and therapeutic strategies, promoting personalized medicine, and advancing scientific research in the field of metabolism and liver diseases.**(International Journal of Biomedicine. 2024;14(3):478-483.)**

Keywords: liver • lipids • choline • magnetic resonance spectroscopy • single volume sequence

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Abbreviations

FLD, fatty liver disease; **LDL-C**, low-density lipoprotein cholesterol; **MRS**, magnetic resonance spectroscopy; **NAFLD**, non-alcoholic fatty liver disease; **ppm**, part-per-million; **SVS**, single volume sequence.

Introduction

The intricate interplay between hepatic choline and lipid metabolism holds significant implications for metabolic health, particularly among young Saudi women. Choline, a vital nutrient, and lipids play pivotal roles in liver function and overall metabolic homeostasis. However, the precise dynamics of choline and lipid metabolism in young females remain understudied in Saudi literature. To address this gap, the present study undertakes an advanced investigation into hepatic choline and lipid profiling among young Saudi women. Leveraging state-of-the-art Magnetic Resonance Spectroscopy (MRS) techniques, this research endeavors to unravel the complex metabolic dynamics underlying liver function and their broader health implications.[1-6]

Choline, an essential nutrient involved in phospholipid synthesis and lipid transport, is integral to liver health. Concurrently, lipids serve as critical energy sources and structural components of cell membranes. Choline is

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intricately involved in various metabolic pathways essential for liver function. Studies have elucidated the role of choline in phospholipid synthesis, bile acid metabolism, and lipid transport. Choline deficiency has been linked to hepatic steatosis, liver dysfunction, and increased risk of metabolic disorders such as non-alcoholic fatty liver disease NAFLD among diverse populations.[7-11]

Lipid profiling, including the assessment of triglycerides, cholesterol, and fatty acids, provides valuable insights into metabolic dysregulation and disease risk. Research has highlighted the association between altered lipid profiles, particularly elevated triglycerides and low-density lipoprotein cholesterol (LDL-C), and increased risk of kidney and cardiovascular disease, and metabolic syndrome. Understanding the nuances of lipid metabolism is crucial for identifying individuals at risk and implementing targeted interventions.[12-17]

Gender and ethnic disparities in metabolic health underscore the importance of studying metabolic dynamics among specific demographic groups, including young Saudi women. Research has revealed unique metabolic profiles and risk factors associated with metabolic disorders in women, compared to men. Additionally, ethnic differences in metabolic phenotype and genetic predisposition contribute to variations in disease susceptibility and treatment response.[18]

Alterations in lipid and choline metabolism can signify metabolic dysregulation and may serve as early markers of metabolic disorders, such as fatty liver disease (FLD), insulin resistance, and cardiovascular disease. Recognizing these metabolic abnormalities allows for early intervention and preventive measures. The correlation between liver lipids and choline levels can inform diagnostic and therapeutic strategies for metabolic disorders. Biomarkers derived from lipid and choline profiling may aid in disease diagnosis and prognosis, and monitoring of treatment efficacy. Understanding individual variations in lipid and choline metabolism enables the development of personalized healthcare approaches. Tailoring interventions based on an individual's metabolic profile enhances treatment outcomes and reduces the risk of adverse effects. Insights into the relationship between lipids and choline levels can guide nutritional strategies to optimize liver health. Promoting dietary habits rich in choline and healthy lipid sources may help prevent or mitigate the risk of metabolic disorders.[10,11,16,19,20]

Magnetic resonance spectroscopy (MRS) offers a non-invasive and highly sensitive method for assessing hepatic choline and lipid levels in vivo. Recent advancements in MRS technology have enabled precise quantification of choline-containing compounds and lipid fractions within the liver. Studies utilizing MRS have provided valuable insights into the pathophysiology of metabolic disorders and the efficacy of therapeutic interventions.[21] MRS enables the visualization of relative tissue metabolite concentrations across a two- or three-dimensional spectrum through the chemical shift phenomenon. In a magnetic resonance spectrum, the signal intensity and frequency of a chemical or metabolite within a specific voxel are plotted. MRS can quantify liver fat by assessing lipid peaks and aid in diagnosing malignancies, typically by evaluating the choline peak. Interpreting MRS data necessitates specialized postprocessing software and is limited by technical factors such as a low signal-to-noise ratio. The application of MRS in liver assessment is an advancing field, offering potential enhancements in the diagnostic precision of tissue characterization when spectra are analyzed alongside magnetic resonance images.[22-25]

Adequate choline intake, coupled with good nutrition and regular physical exercise, plays a pivotal role in ensuring healthy liver function and mitigating the risk of FLD. Incorporating choline-rich foods, such as eggs, meat, fish, and certain vegetables, into one's diet provides the necessary substrate for optimal liver function. Moreover, adopting a balanced diet rich in fruits, vegetables, whole grains, and lean proteins not only supports choline intake but also promotes overall liver health by supplying essential vitamins, minerals, and antioxidants. Complementing proper nutrition with regular physical exercise further enhances liver function by improving insulin sensitivity, promoting lipid utilization, and reducing hepatic fat accumulation. Physical activity stimulates liver metabolism, facilitating the breakdown and clearance of lipids from the liver, thus reducing the FLD risk. By synergistically combining choline intake, nutritious eating habits, and routine exercise, individuals can safeguard their liver health, ward off FLD, and foster overall well-being.[19,20,22,26-30]

In summary, this study underscores the importance of advanced assessment techniques, such as MRS, in unraveling metabolic dynamics and their implications for health among young Saudi women. By integrating knowledge from choline metabolism, lipid profiling, and ethnic-specific considerations (Saudi female volunteers), this research aims to contribute to the understanding of metabolic health and facilitate personalized healthcare strategies tailored to the needs of this demographic. The investigation of the relationship between lipids and choline levels in the liver holds significant importance for several reasons, such as understanding liver health because the liver is a vital organ responsible for numerous metabolic functions, including lipid metabolism and choline utilization. By elucidating the correlation between lipids and choline levels, researchers gain insights into the mechanisms underlying liver health and function.

Significance of the Study and Rationale for It

Recent years have witnessed notable shifts in lifestyle patterns among young adults aged 18 to 25 in Saudi Arabia, leading to an increase in the consumption of fatty products, soft drinks, and caffeinated beverages; these dietary choices affect amino acids and lipid levels in the liver. This study aims to investigate the correlation between liver choline and lipid levels among university students. Such research serves as a foundational step in raising awareness and promoting health education regarding the significant association between lifestyle and liver lipids and choline fatty diseases.

Materials and Methods

This cross-sectional research explored the relationship between liver choline and fat levels, employing MRS to estimate these levels. A total of 29 healthy female students from

the radiological sciences department, aged between 18 and 25 years, participated in the study. MRS scans were conducted using a 1.5 tesla Philips scanner using single volume sequence (SVS)[12,14,21,23] with an echo time (TE) of 30 ms, and repetition time (TR) of 1500 ms, resulting in the liver choline and lipid peaks depicted in Figure 1. Data confidentiality was ensured, including collected estimated lipids and choline levels from the magnetic resonance spectra, which tabulated in an Excel spreadsheet for all participants.

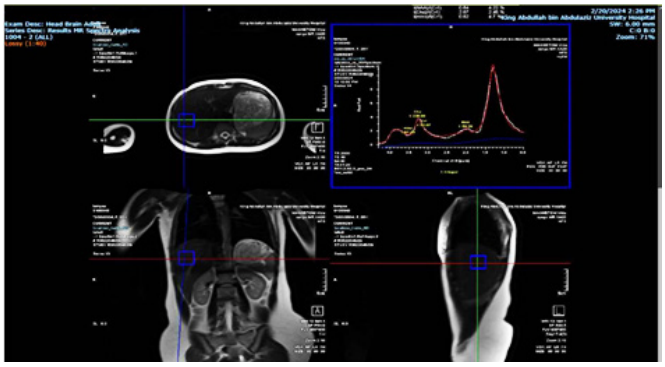

Fig. 1. *1.5 tesla Philips scanner liver MR image and the corresponding spectrum, SVS with TE=30 ms and TR=1500 ms.*

Statistical analysis was performed using the statistical software package SPSS version 20.0 (SPSS Inc, Armonk, NY: IBM Corp). Baseline characteristics were summarized as frequencies and percentages for categorical variables and mean ± standard deviation (SD) for continuous variables. Pearson's Correlation Coefficient (r) was used to determine the strength of the relationship between the two continuous variables. A probability value of $P<0.05$ was considered statistically significant.

Results

MRS offers exceptional visibility in detecting liver lipids and choline levels, providing valuable insights into hepatic metabolism and health. This non-invasive technique allows for the precise measurement of lipid and choline concentrations within the liver tissue, facilitating the early detection of metabolic abnormalities and liver pathologies. By analyzing the spectral patterns of lipids and choline, MRS enables clinicians to assess liver health without the need for invasive procedures, offering a safe and reliable means of evaluating hepatic function and guiding clinical management decisions.

In this section, we disclose the outcomes of MRS liver spectra alongside the associated choline and lipid measurements within the volunteers' livers and correlate the levels of choline and lipids in the liver using the Pearson correlation test.

Liver Lipids and Choline Spectrum

Figure 1 displays the outcomes of the SVS spectroscopy sequence used in this study, indicating the presence of lipid peaks within the frequency range of 0.9-1.2 ppm and choline peaks at 3.2 ppm. The MRI images highlight the pixels corresponding to the choline spectrum and the estimated choline levels.

Liver Lipids and Choline Levels Correlation Analysis Test

Table 1 outlines the liver lipid levels, estimated through MRS for the healthy young female volunteers, as part of a descriptive analysis of the lipids (0.13659±0.188526) and choline (0.08279±0.191522). The Pearson correlation test revealed a statistically significant association between choline and lipid levels in the liver (r=0.367, P=0.025) (Table 2).

Table 1.

Liver choline and lipids descriptive analysis.

	Mean	SD	N
Lipids	0.13659	0.188526	29
Choline	0.08279	0.191522	29

Table 2.

Pearson correlation between liver choline and lipids.

		Lipids	Choline
Lipids	Pearson Correlation	1	0.367
	Sig. (1-tailed)		0.025
	N	29	29
Choline	Pearson Correlation	0.367	1
	Sig. (1-tailed)	0.025	
	N	29	29

Discussion

This research illustrates the effectiveness of MRS in identifying low levels of choline and lipids in healthy adult females. The choline peak is prominently observed at 3.2 ppm, while the lipid peak consistently appears within the frequency range of 0.9-1.2 ppm for all participants. This underscores MRS's ability to detect trace amounts of amino acids like choline and liver lipids. The observed spectrum and the visibility of liver choline and lipid peaks align closely with findings from prior studies on MRS of liver lipids and amino acids.[12,14,21,23]

The study data were tabulated, and descriptive analysis was done for the estimated liver lipids and choline levels. Pearson correlation test results suggest a statistically significant association between choline and lipid concentrations in the liver (r=0.367, P=0.025), emphasizing the interdependence of these two crucial components in hepatic metabolism. Such findings underscore the importance of considering the intricate relationship between choline and lipids for a comprehensive understanding of liver health and metabolic function.

Choline supports liver function by maintaining cell membranes, aiding fat metabolism, and participating in critical biochemical reactions. A balanced diet rich in choline

contributes to overall liver health. For more than 50 years, researchers have acknowledged the association between choline deficiency and the buildup of hepatic lipids.[31] This understanding has led to the creation of choline-deficient diets as a means to induce models of NAFLD in animal studies. In human populations, NAFLD stands as the most dominant liver condition globally, impacting approximately 30% of Western and 17% of Eastern populations.[32]

Previous studies by Sherriff et al. [33] and Sha et al.[5] have indicated that inadequate choline levels and insufficient dietary intake elevate the likelihood of developing NAFLD. This suggests that low choline concentration could significantly contribute to the heightened risk of NAFLD in individuals with suboptimal choline intake from their daily diet. Furthermore, this association may potentially aggravate the risk of cardiovascular disease among those affected by NAFLD. The study presented here demonstrates a correlation between choline and liver lipids in young healthy females. This finding aligns with these studies, suggesting that normal liver choline levels are associated with balanced lipid levels in healthy young individuals.

A previous study conducted by Ficher et al.[18] demonstrated that both sex and age increase the likelihood of developing FLD when one is deprived of dietary choline. In Ficher's study, 77% of men and 80% of postmenopausal women developed fatty liver or muscle damage, whereas only 44% of premenopausal women exhibited such signs of organ dysfunction. The volunteers recruited for the current study are exclusively premenopausal females who exhibited normal choline levels, and no indication of increased liver fat in the MRS liver spectrum. Although the sex dependency aspect cannot be explored in this study due to the inclusion of only females, the findings from Ficher's study, coupled with the results of the current investigation, suggest that young females may have a reduced risk of developing fatty liver issues by adhering to a healthy diet.

Studies by Fischbach & Bruhn,[34] Di Martino et al.,[35] Caussy et al.,[36] and Jones[37] have contributed significantly to the understanding of MRS in assessing liver choline and lipid levels. Their research has demonstrated the efficacy of MRS in accurately estimating choline and lipid concentrations, as well as establishing correlations between these metabolites. Moreover, their findings have underscored the association between low choline levels and the development of FLD.

The current study's results align with these earlier investigations, as evidenced by the clear separation of choline and lipid peaks in the spectra of all volunteers. This observation validates the robustness of MRS in discerning between these metabolites, consistent with prior research findings. Furthermore, the utilization of data analysis software in the present study has facilitated accurate estimation of choline and lipid levels, mirroring the outcomes reported in previous studies.

In summary, the collective body of research, including the present study, emphasizes the utility of MRS in quantifying liver choline and lipid concentrations. These findings not only contribute to our understanding of liver metabolism but also have implications for the diagnosis and management of FLD.

The correlation between dietary choices, liver health, and the development of FLD underscores the importance of promoting balanced and nutritious eating habits among young generations. Educating individuals about the significance of dietary nutrients, including choline and healthy lipid sources, can help mitigate the risk of metabolic disorders and promote liver health. By making informed dietary choices, young individuals can safeguard their liver function and reduce the likelihood of developing serious diseases, ultimately supporting their overall health and well-being.

This study's implications extend beyond mere academic interest; they bear profound implications for promoting the well-being of young Saudi females. By clarifying the complex interplay between liver lipids and choline levels, researchers can tailor preventive measures and treatment modalities specific to this demographic, thus enhancing their overall health outcomes and quality of life. Furthermore, such investigations contribute to the broader landscape of medical research, facilitating advancements in our understanding of liver physiology and metabolic disorders, which in turn can inform the development of innovative therapies and public health initiatives tailored to the needs of young Saudi women.

Conclusion

Research into the correlation between liver lipids and choline levels contributes to the advancement of scientific knowledge in the fields of hepatology, nutrition, and metabolic medicine. By uncovering novel mechanisms and pathways, this research paves the way for innovative approaches to disease prevention and treatment. The dietary preferences of young generations have a profound impact on the levels of crucial nutrients such as choline and lipids in the liver, ultimately influencing their overall health status. The inadequate intake or imbalance of essential nutrients can disrupt metabolic processes, leading to adverse health outcomes, including the development of serious conditions such as FLD. Investigating the relationship between liver lipids and choline levels is crucial for understanding liver health, identifying metabolic dysregulation, informing diagnostic and therapeutic strategies, promoting personalized medicine, and advancing scientific research in the field of metabolism and liver diseases.

Ethical Considerations

The research took place on the university campus and at the medical imaging department of King Abdullah Bin Abdulaziz University Hospital (KAAUH) during the second term of the 2022-2023 academic year, following the receipt of ethical approval (IRB) from the research committee of Princess Nourah bint Abdulrahman University (PNU). The IRB registration number with KACST, KSA is HAP-01-R-059, November 29, 2022, and the IRB Log Number is 22-1005.

Competing Interests

The author declares that there is no conflict of interest.

References

1. Canty DJ, Zeisel SH. Lecithin and choline in human health and disease. Nutr Rev. 1994 Oct;52(10):327-39. doi: 10.1111/j.1753-4887.1994.tb01357.x. PMID: 7816350.
2. Mehedint MG, Zeisel SH. Choline's role in maintaining liver function: new evidence for epigenetic mechanisms. Curr Opin Clin Nutr Metab Care. 2013 May;16(3):339-45. doi: 10.1097/MCO.0b013e3283600d46. PMID: 23493015; PMCID: PMC3729018.
3. Zeisel SH. Choline: critical role during fetal development and dietary requirements in adults. Annu Rev Nutr. 2006;26:229-50. doi: 10.1146/annurev.nutr.26.061505.111156. PMID: 16848706; PMCID: PMC2441939.
4. Cho E, Willett WC, Colditz GA, Fuchs CS, Wu K, Chan AT, Zeisel SH, Giovannucci EL. Dietary choline and betaine and the risk of distal colorectal adenoma in women. J Natl Cancer Inst. 2007 Aug 15;99(16):1224-31. doi: 10.1093/jnci/djm082. Epub 2007 Aug 8. PMID: 17686825; PMCID: PMC2441932.
5. Sha W, da Costa KA, Fischer LM, Milburn MV, Lawton KA, Berger A, Jia W, Zeisel SH. Metabolomic profiling can predict which humans will develop liver dysfunction when deprived of dietary choline. FASEB J. 2010 Aug;24(8):2962-75. doi: 10.1096/fj.09-154054. Epub 2010 Apr 6. PMID: 20371621; PMCID: PMC2909293.
6. Öz G, Deelchand DK, Wijnen JP, Mlynárik V, Xin L, Mekle R, Noeske R, Scheenen TWJ, Tkáč I; Experts' Working Group on Advanced Single Voxel 1H MRS. Advanced single voxel ^{1}H magnetic resonance spectroscopy techniques in humans: Experts› consensus recommendations. NMR Biomed. 2020 Jan 10:e4236. doi: 10.1002/nbm.4236. Epub ahead of print. PMID: 31922301; PMCID: PMC7347431.
7. Zhu J, Wu Y, Tang Q, Leng Y, Cai W. The effects of choline on hepatic lipid metabolism, mitochondrial function and antioxidative status in human hepatic C3A cells exposed to excessive energy substrates. Nutrients. 2014 Jul 9;6(7):2552-71. doi: 10.3390/nu6072552. PMID: 25010553; PMCID: PMC4113756.
8. Thomas EL, Hamilton G, Patel N, O'Dwyer R, Doré CJ, Goldin RD, Bell JD, Taylor-Robinson SD. Hepatic triglyceride content and its relation to body adiposity: a magnetic resonance imaging and proton magnetic resonance spectroscopy study. Gut. 2005 Jan;54(1):122-7. doi: 10.1136/gut.2003.036566. PMID: 15591516; PMCID: PMC1774370.
9. Cho SG, Kim MY, Kim HJ, Kim YS, Choi W, Shin SH, Hong KC, Kim YB, Lee JH, Suh CH. Chronic hepatitis: in vivo proton MR spectroscopic evaluation of the liver and correlation with histopathologic findings. Radiology. 2001 Dec;221(3):740-6. doi: 10.1148/radiol.2213010106. PMID: 11719670.
10. Corbin KD, Zeisel SH. Choline metabolism provides novel insights into nonalcoholic fatty liver disease and its progression. Curr Opin Gastroenterol. 2012 Mar;28(2):159-65. doi: 10.1097/MOG.0b013e32834e7b4b. PMID: 22134222; PMCID: PMC3601486.
11. Teodoro JS, Rolo AP, Duarte FV, Simões AM, Palmeira CM. Differential alterations in mitochondrial function induced by a choline-deficient diet: understanding fatty liver disease progression. Mitochondrion. 2008 Dec;8(5-6):367-76. doi: 10.1016/j.mito.2008.07.008. Epub 2008 Aug 15. PMID: 18765303.
12. Szczepaniak LS, Nurenberg P, Leonard D, Browning JD, Reingold JS, Grundy S, Hobbs HH, Dobbins RL. Magnetic resonance spectroscopy to measure hepatic triglyceride content: prevalence of hepatic steatosis in the general population. Am J Physiol Endocrinol Metab. 2005 Feb;288(2):E462-8. doi: 10.1152/ajpendo.00064.2004. Epub 2004 Aug 31. PMID: 15339742.
13. Younossi ZM, Koenig AB, Abdelatif D, Fazel Y, Henry L, Wymer M. Global epidemiology of nonalcoholic fatty liver disease-Meta-analytic assessment of prevalence, incidence, and outcomes. Hepatology. 2016 Jul;64(1):73-84. doi: 10.1002/hep.28431. Epub 2016 Feb 22. PMID: 26707365.
14. Qi YM, Xiao EH. Advances in application of novel magnetic resonance imaging technologies in liver disease diagnosis. World J Gastroenterol. 2023 Jul 28;29(28):4384-4396. doi: 10.3748/wjg.v29.i28.4384. PMID: 37576700; PMCID: PMC10415971.
15. Abbate M, Mascaró CM, Montemayor S, Casares M, Gómez C, Ugarriza L, Tejada S, Abete I, Zulet MA, Sureda A, Martínez JA, Tur JA. Non-Alcoholic Fatty Liver Disease Is Associated with Kidney Glomerular Hyperfiltration in Adults with Metabolic Syndrome. J Clin Med. 2021 Apr 16;10(8):1717. doi: 10.3390/jcm10081717. PMID: 33923418; PMCID: PMC8073467.
16. Raubenheimer PJ, Nyirenda MJ, Walker BR. A choline-deficient diet exacerbates fatty liver but attenuates insulin resistance and glucose intolerance in mice fed a high-fat diet. Diabetes. 2006 Jul;55(7):2015-20. doi: 10.2337/db06-0097. PMID: 16804070.
17. Yu D, Shu XO, Xiang YB, Li H, Yang G, Gao YT, Zheng W, Zhang X. Higher dietary choline intake is associated with lower risk of nonalcoholic fatty liver in normal-weight Chinese women. J Nutr. 2014 Dec;144(12):2034-40. doi: 10.3945/jn.114.197533. Epub 2014 Oct 15. PMID: 25320186; PMCID: PMC4230213.
18. Fischer LM, daCosta KA, Kwock L, Stewart PW, Lu TS, Stabler SP, Allen RH, Zeisel SH. Sex and menopausal status influence human dietary requirements for the nutrient choline. Am J Clin Nutr. 2007 May;85(5):1275-85. doi: 10.1093/ajcn/85.5.1275. PMID: 17490963; PMCID: PMC2435503.
19. Młodzik-Czyżewska M, Malinowska A, Chmurzyńska A. Associations between choline intake, body composition, lipid profile, and liver status in healthy adults. Proc Nutr Soc. 2020;79(OCE2). doi:10.1017/s0029665120003195.
20. Chai C, Chen L, Deng MG, Liang Y, Liu F, Nie JQ. Dietary choline intake and non-alcoholic fatty liver disease (NAFLD) in U.S. adults: National Health and Nutrition Examination Survey (NHANES) 2017-2018. Eur J Clin Nutr. 2023 Dec;77(12):1160-1166. doi: 10.1038/s41430-023-01336-1. Epub 2023 Aug 26. PMID: 37634048.
21. Lăpădat AM, Florescu LM, Manea NC, Gheonea DI, Pirici D, Tudoraşcu DR, Ene R, Gheonea IA. MR spectroscopy of the liver - a reliable non-invasive alternative for evaluating non-alcoholic fatty liver disease. Rom J Morphol Embryol. 2020;61(1):73-80. doi: 10.47162/RJME.61.1.08. PMID: 32747897; PMCID: PMC7728118.
22. Zeisel SH, da Costa KA. Choline: an essential nutrient for public health. Nutr Rev. 2009 Nov;67(11):615-23. doi: 10.1111/j.1753-4887.2009.00246.x. PMID: 19906248; PMCID: PMC2782876.

23. Xu L, Liu B, Huang Y, Liu X, Zhang SW, Xin XG, Zheng JZ. 3.0 T proton magnetic resonance spectroscopy of the liver: quantification of choline. World J Gastroenterol. 2013 Mar 7;19(9):1472-7. doi: 10.3748/wjg.v19.i9.1472. PMID: 23539666; PMCID: PMC3602508.
24. Müller C, Hübner F, Bisdas S, Herzog C, Hammerstingl RM, Ackermann H, Vorbuchner M, Vogl TJ. In-vivo-Protonenspektroskopie des Leberparenchyms: Technik und Ergebnisse [In vivo proton MR spectroscopy of normal liver parenchyma: technique and results]. Rofo. 2006 Nov;178(11):1128-36. German. doi: 10.1055/s-2006-927136.
25. Qayyum A. MR spectroscopy of the liver: principles and clinical applications. Radiographics. 2009 Oct;29(6):1653-64. doi: 10.1148/rg.296095520.
26. Romero-Gómez M, Zelber-Sagi S, Trenell M. Treatment of NAFLD with diet, physical activity and exercise. J Hepatol. 2017 Oct;67(4):829-846. doi: 10.1016/j.jhep.2017.05.016. Epub 2017 May 23. PMID: 28545937.
27. Chang TY, Wu CH, Chang CY, Lee FJ, Wang BW, Doong JY, Lin YS, Kuo CS, Huang RS. Optimal Dietary Intake Composition of Choline and Betaine Is Associated with Minimized Visceral Obesity-Related Hepatic Steatosis in a Case-Control Study. Nutrients. 2022 Jan 8;14(2):261. doi: 10.3390/nu14020261.
28. Fabbrini E, Sullivan S, Klein S. Obesity and nonalcoholic fatty liver disease: biochemical, metabolic, and clinical implications. Hepatology. 2010 Feb;51(2):679-89. doi: 10.1002/hep.23280. PMID: 20041406; PMCID: PMC3575093.
29. Younossi ZM. Non-alcoholic fatty liver disease - A global public health perspective. J Hepatol. 2019 Mar;70(3):531-544. doi: 10.1016/j.jhep.2018.10.033. Epub 2018 Nov 9. PMID: 30414863.
30. Zeisel SH. Metabolic crosstalk between choline/1-carbon metabolism and energy homeostasis. Clin Chem Lab Med. 2013 Mar 1;51(3):467-75. doi: 10.1515/cclm-2012-0518.
31. Nakamura T, Nakamura S, Karoji N, Aikawa T, Suzuki O. Hepatic function tests in heavy drinkers among workmen. Tohoku J Exp Med. 1967 Nov;93(3):219-26. doi: 10.1620/tjem.93.219. PMID: 5587662.
32. Han SK, Baik SK, Kim MY. Non-alcoholic fatty liver disease: Definition and subtypes. Clin Mol Hepatol. 2023 Feb;29(suppl):S5-S16. doi: 10.3350/cmh.2022.0424. Epub 2022 Dec 28. PMID: 36577427; PMCID: PMC10029964.
33. Sherriff JL, O'Sullivan TA, Properzi C, Oddo JL, Adams LA. Choline, Its Potential Role in Nonalcoholic Fatty Liver Disease, and the Case for Human and Bacterial Genes. Adv Nutr. 2016 Jan 15;7(1):5-13. doi: 10.3945/an.114.007955.
34. Fischbach F, Bruhn H. Assessment of in vivo 1H magnetic resonance spectroscopy in the liver: a review. Liver Int. 2008 Mar;28(3):297-307. doi: 10.1111/j.1478-3231.2007.01647.x.
35. Di Martino M, Pacifico L, Bezzi M, Di Miscio R, Sacconi B, Chiesa C, Catalano C. Comparison of magnetic resonance spectroscopy, proton density fat fraction and histological analysis in the quantification of liver steatosis in children and adolescents. World J Gastroenterol. 2016 Oct 21;22(39):8812-8819. doi: 10.3748/wjg.v22.i39.8812. PMID: 27818597; PMCID: PMC5075556.
36. Caussy C, Reeder SB, Sirlin CB, Loomba R. Noninvasive, Quantitative Assessment of Liver Fat by MRI-PDFF as an Endpoint in NASH Trials. Hepatology. 2018 Aug;68(2):763-772. doi: 10.1002/hep.29797.
37. Jones JG. Non-Invasive Analysis of Human Liver Metabolism by Magnetic Resonance Spectroscopy. Metabolites. 2021 Oct 29;11(11):751. doi: 10.3390/metabo11110751. PMID: 34822409; PMCID: PMC8623827.