Fructose, high-fructose corn syrup, sucrose, and nonalcoholic fatty liver disease or indexes of liver health: a systematic review and meta-analysis¹⁻⁴

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ABSTRACT
Background: Concerns have been raised about the concurrent temporal trend between simple sugar intakes, especially of fructose or high-fructose corn syrup (HFCS), and rates of nonalcoholic fatty liver disease (NAFLD) in the United States.

Objective: We examined the effect of different amounts and forms of dietary fructose on the incidence or prevalence of NAFLD and indexes of liver health in humans.

Design: We conducted a systematic review of English-language, human studies of any design in children and adults with low to no alcohol intake and that reported at least one predetermined measure of liver health. The strength of the evidence was evaluated by considering risk of bias, consistency, directness, and precision.

Results: Six observational studies and 21 intervention studies met the inclusion criteria. The overall strength of evidence for observational studies was rated insufficient because of high risk of biases and inconsistent study findings. Of 21 intervention studies, 19 studies were in adults without NAFLD (predominantly healthy, young men) and 1 study each in adults or children with NAFLD. We found a low level of evidence that a hypercaloric fructose diet (supplemented by pure fructose) increases liver fat and aspartate aminotransferase (AST) concentrations in healthy men compared with the maintenance diet. In addition, there was a low level of evidence that hypercaloric fructose and glucose diets have similar effects on liver fat and indexes of liver health in humans.

Conclusions: On the basis of indirect comparisons across study findings, the apparent association between indexes of liver health (ie, liver fat, hepatic de novo lipogenesis, alanine aminotransferase, AST, and γ-glutamyl transpeptidase) and fructose or sucrose intake appear to be confounded by excessive energy intake. Overall, the available evidence is not sufficiently robust to draw conclusions regarding effects of fructose, HFCS, or sucrose consumption on NAFLD. Am J Clin Nutr 2014;100:833–49.

INTRODUCTION
Nonalcoholic fatty liver disease (NAFLD)⁵ is characterized by hepatic steatosis, which is the abnormal triglyceride accumulation in liver cells. It has been suggested that NAFLD should be recognized as the hepatic component of metabolic syndrome (1). NAFLD was formerly called nonalcoholic steatohepatitis (NASH), which was defined as steatosis with inflammation and hepatocyte necrosis. More recently, clinical practice guidelines have defined NAFLD as hepatic steatosis that is not caused by significant alcohol consumption, other competing causes such as the use of a steatogenic medication or genetic disorders, and coexisting causes of chronic liver disease such as Wilson’s disease (2).

NAFLD now refers to a spectrum of pathologic disorders that range from simple steatosis and steatohepatitis to advanced fibrosis and cirrhosis (2, 3). The cause, pathophysiology, and pathogenesis of NAFLD and NASH remain poorly understood (4, 5).

Lipid accumulation in hepatocytes may lead to oxidative stress in the endoplasmic reticulum and mitochondria, which can promote inflammation in the liver (6, 7). An assessment of hepatic steatosis requires either radiologic measures (eg, ultrasound, computed tomography, magnetic resonance imaging, proton magnetic resonance spectroscopy, or transient elastography) or a liver biopsy to provide histologic evidence of triglyceride accumulation. Excessive triglyceride accumulation is defined as >5% of hepatocytes that contained detectable triglyceride (4, 8). Some studies have used a combination of noninvasive measures, including BMI, waist circumference, plasma triglyceride concentrations, and liver enzymes as indicators for the potential diagnosis of NAFLD (9, 10). There is currently no consensus for a noninvasive reference standard for the diagnosis of NAFLD.

A systematic review of published studies related to NAFLD...
prevalence and incidence concluded that definitions for the diagnosis of NAFLD are heterogeneous and cautioned that the estimated prevalence varies on the basis of diagnostic technique (11). Despite these caveats, there appears to be an increasing trend in the prevalence of NAFLD in adults and adolescents living in the United States over the past 20 y (12, 13).

It has been suggested that there is a causative relation between the increased prevalence of NAFLD and related disorders (ie, obesity, diabetes, cardiovascular disease, hypertension, cancer and metabolic syndrome) and intakes of sweeteners, particularly fructose (14). The “fructose hypothesis” in part has been driven by data from animal studies and in part by historical trends (15, 16). Specifically, animal studies have shown that high-fructose, compared with glucose, diets result in an increased hepatic triglyceride content (17–21). Of note, although they are useful for elucidating the potential mechanisms for the development and progression of NAFLD, animal models cannot confirm the cause and pathophysiology in humans. Historical dietary consumption trends have suggested an 18% increase in daily energy intake between the 1977–1978 Nationwide Food Consumption Survey and the 1999–2004 NHANES (22). Mean total fructose intake as a percentage of carbohydrate followed a similar trend pattern. More recent NHANES data have suggested a decreasing trend in the average per capita and population mean energy intake of added sugar (14, 23).

The aim of this study was to perform a systematic literature review to assess data related to fructose intake as a monoglyceride or diglyceride and indexes of liver health in humans. Also examined was the potential modifying factors for any associations if identified.

METHODS

In this systematic review, we defined dietary fructose to include the monosaccharide forms high-fructose corn syrup (HFCS) and honey and the disaccharide form sucrose. We formulated initial key questions on the basis of a generic analytic framework for assessments of nutrient and outcome relations (24).

We presented the initial key questions and study eligibility criteria to a technical expert panel (TEP) who served in an advisory capacity to help refine questions, review literature search terms and strategies, identify potential confounding factors, and appraise variables for the review of evidence. During the review process, we consulted the TEP regarding technical details (eg, refinements of quality appraisal items and study eligibility criteria). Neither the sponsor nor TEP members participated in our research meetings or reviewed or synthesized the evidence.

We followed the methods for conducting a systematic review outlined in the Institute of Medicine’s Standards for Systematic Reviews (25) with the exception of small modifications as indicated. We reported our study results according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement (26).

Data sources and searches

We conducted literature searches of studies in Ovid MEDLINE (1946 to March 2014 week 2), the Cochrane Central Register of Controlled Trials (through January 2014), CAB Abstracts (1973–2014 week 11), and Global Health (1910–2014 week 11) databases (gateway.ovid.com). All studies published in the English language with human subjects were screened to identify articles relevant to our key questions. Our search strategy used the National Library of Medicine’s Medical Subject Headings (MeSH) keyword nomenclature developed for MEDLINE. See Supplemental Table 1 under “Supplemental data” in the online issue for a description of the full search strategy. The search strategy combined MeSH or search terms for fructose, sucrose, sweetening agents, and selected sugar-rich foods with MeSH or search terms for fatty liver, hepatic triglycerides, diagnostic techniques for NAFLD, and liver enzymes. We confirmed that our search strategy identified key articles provided by the TEP. We also screened reference lists of selected reviews and primary articles for additional publications. We did not search for unpublished studies or articles in the gray literature.

Study selection and eligibility criteria

All abstracts identified through the literature search were screened independently by ≥2 investigators on the basis of predetermined eligibility criteria with a low threshold to exclude irrelevant abstracts such as animal or in vitro studies and studies that did not investigate diet and disease associations. Full-text articles were retrieved for abstracts that were accepted by at least one investigator. Articles were evaluated independently by teams of 2 investigators for eligibility. Disagreement on eligibility was resolved in consultation with a third team member. In addition, we screened full-text articles of included studies in published systematic reviews and meta-analyses that investigated effects of fructose on body weight (27), uric acid (28), glycemic effects (29), blood pressure (30), and blood lipids (31) for our outcomes of interest.

We included studies of any design in adults and children (≥4 y of age) with low (as defined by original articles) to no alcohol intake. Our interventions or exposures of interest were free (pure) fructose, total fructose, sucrose, HFCS, and sugar-sweetened beverages (if the absolute amount of fructose or sucrose was quantified and reported in the original article). The term “(total) fructose intake”, which is commonly used in epidemiologic studies, refers to fructose consumption from all sources or forms. Outcomes of interests were an NAFLD or NASH diagnosis as defined by original studies and predetermined indexes of liver health including intrahepatocellular lipids (IHCLs) (liver fat), hepatic de novo lipogenesis (DNL), liver enzymes [alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase, and γ-glutamyl transpeptidase (GGT)], additional indicators of liver function (eg, bilirubin), and liver fibrosis markers (eg, collagen IV or procollagen III). We excluded studies of patients with fructose intolerance, infection-related liver disease, metabolic diseases that affect the liver, Wilson’s disease, cirrhosis, and hepatocellular carcinoma. We also excluded studies that provided fructose intravenously, investigated oligofructose (not our interventions or exposures of interest), or did not report the fructose, sucrose, or HFCS dose. Because of a scarcity of data, we did not exclude studies on the basis of a small sample size or short study duration although these factors were taken into consideration when we evaluated the strength of the body of the evidence.

Data extraction and quality assessment

Data were extracted by using standardized forms, and all extracted data are available at the Systematic Review Data Repository (http://srdr.ahrq.gov/projects/64). We extracted information on study characteristics, baseline population characteristics, background diet, dietary assessment methods for fructose or sucrose intake, interventions (for observational studies only), confounders and effect modifiers that were adjusted
for in statistical analysis, biological outcomes, and information used to assess the risk of bias (ROB).

We assessed the ROB (or methodologic quality) for each individual study by using standardized assessment instruments that were modified to include nutrition-specific quality items (32). See Supplemental Table 2 under “Supplemental data” in the online issue for a description of details of the ROB assessment for each study. Briefly, we rated each study as being of high, medium, or low ROB by using a modified Cochrane ROB tool (33) for controlled intervention studies, which was used previously in a systematic review for Dietary Reference Intake values (34), and a modified Newcastle-Ottawa scale (35) for observational studies. We consulted the TEP on methodologic quality items and added 2 quality items on dietary assessment methods to the Newcastle-Ottawa scale (35). The grading was outcome-specific such that a given study that reported its primary outcome well but conducted an incomplete analysis of a secondary outcome was graded as having different qualities for the 2 outcomes. The quality assessment was performed by an investigator and confirmed by another investigator. Disagreement was resolved in consultation with a third team member.

Data synthesis

We qualitatively synthesized all included studies in summary tables by considering their study design, population characteristics, sources and forms of dietary fructose, energy balance of the dietary fructose or sucrose intervention and its comparison, background diet, and types of outcome measure or definitions. Studies were first grouped by the fructose exposure forms, monosaccharide fructose (including pure fructose sweeteners, honey, and HFCS) or sucrose (1:1 fructose-glucose disaccharide), and results were evaluated separately on the basis of the overall energy balance (hypercaloric, hypocaloric, or isocaloric) of diets and on the energy balance of the sugar intervention and its comparison (a positive, neutral, or negative comparison). The term isocaloric diet is used to describe a weight-maintenance diet and hypercaloric and hypocaloric diets were used to describe diets that resulted in a weight gain or weight lost, respectively. A neutral comparison was defined as equivalent energy content between the sugar intervention and its comparison intervention, and a positive or negative comparison was defined as a higher or lower energy content in the sugar intervention than its comparison intervention, respectively. We evaluated the strength of the body of evidence for each exposure-comparison pair within each outcome of interest with respect to the following 4 domains: ROB, consistency, directness, and precision (36). The strength of the body of evidence rating (ie, insufficient, low, moderate, or high level of evidence) was based on the consensus of all team investigators. When only one study was available for each exposure-comparison pair within each outcome of interest, we rated the strength of the body of evidence as insufficient.

We graphically presented quantitative data by using weighted scatterplots to summarize sample sizes, the exposure-comparison pair, study designs, and specific indexes of liver health investigated in the studies that we reviewed. This approach aimed to provide investigators with information about the type and amount of research available, characteristics of that research, and topics on which sufficient evidence had accumulated for synthesis (37). In addition, when more than 2 controlled intervention studies in the same exposure-comparison pair reported sufficient quantitative data on the same outcome, we pooled study results by using the DerSimonian-Laird random-effects model meta-analysis (38). The effect size and its variance for crossover trials were calculated according to methods recommended in the Cochrane Handbook (39). Briefly, effect sizes were calculated as the difference between sugar-intervention and comparison groups at the end of each intervention. The SD of the mean differences (SD_{diff}) was calculated by using SDs for each intervention (SD_{E} = sugar intervention; SD_{C} = comparison) and an imputed correlation coefficient (Corr) for prepost measurements by using this formula:

\[ SD_{diff} = \sqrt{(SD_{E}^2 + SD_{C}^2 - 2 \times Corr \times SD_{E} \times SD_{C})} \quad (I) \]

An imputed correlation (Corr) value of 0.50 was used to provide a conservative estimate on the basis of the assumption that this value would minimize the error of effect-size estimates. When studies reported data in figures that were not provided numerically, we extracted data points from figures with DataThief III software (www.datathief.org). We tested for heterogeneity with the Cochran Q statistic (considered significant when the P value was less than 0.10) and quantified the extent of heterogeneity with the I^2 index. We defined low, moderate, and high heterogeneity as I^2 values of 25%, 50%, and 75%, respectively. These cutoffs were arbitrary and used for descriptive purposes only (40).

Analyses were conducted with Stata SE 12 software (Stata Corp). All P values were 2 tailed, and a P value less than 0.05 was considered to indicate a statistically significant difference.

RESULTS

Search results

Our literature search yielded 2137 citations of which 104 citations were considered potentially eligible and retrieved in full text (Figure 1). After full-text screening, 27 publications met our study eligibility criteria. Two additional articles were identified from reference lists of published systematic reviews. In total, 27 unique studies in 29 publications were included in this systematic review (41–69).

Observational studies

Three case-control studies (41–43) and one prospective cohort study (66) in adults and 2 cross-sectional studies in children and adolescents (44, 45) were included. Characteristics of these 6 observational studies are summarized in Table 1. We did not conduct the meta-analysis to pool results from the 3 case-control studies because each study only reported unadjusted or underadjusted estimates. Thus, the pooling of unadjusted estimates would have aggregated the confounding bias in studies and may have led to an inappropriate interpretation of meta-analysis results. Findings from the observational studies are qualitatively synthesized in Table 2.

Three case-control studies (all rated at high ROB) compared dietary fructose intakes in a total of 81 adult NAFLD case patients and 40 hospital control patients without NAFLD (41–43). NAFLD was defined by a liver biopsy in 2 studies (41, 42) and by ultrasound and blood variables in the third study (43). One study compared the daily consumption of HFCS- or sugar-containing beverages (kcal/d) in NAFLD cases with that in age-sex-BMI–matched controls (41). The other 2 studies performed unadjusted analyses by comparing total fructose or sucrose intakes (g/d) between NAFLD cases and control patients (42, 43). All 3
case-control studies showed higher mean daily fructose (from all sources, including sugar-sweetened beverages) or sucrose intakes in NAFLD patients compared with hospital control patients. However, the validity of these findings was questionable because of potential biases (eg, recall and selection biases) and unadjusted confounding. One prospective cohort study (low ROB) examined associations between carbohydrate quality (the glycemic index and intakes of sugar, starch, and fiber) and liver enzymes in 866 older adults. The study showed that sugar intake (percentage of total carbohydrate intake) was not associated with ALT or GGT in multivariable, concurrent-change analyses from baseline to 5 y of follow-up (66).

One cross-sectional study (rated at medium ROB) examined associations between dietary sucrose intake and the hepatic fat fraction (HFF) measured by using liver magnetic resonance imaging in 153 overweight or obese children and adolescents with Hispanic ancestry who were living in the United States (44). The study showed that sugar intake (percentage of total carbohydrate intake) was not associated with ALT or GGT in multivariable, concurrent-change analyses from baseline to 5 y of follow-up (66).

One cross-sectional study (rated at medium ROB) examined associations between dietary sucrose intake and the hepatic fat fraction (HFF) measured by using liver magnetic resonance imaging in 153 overweight or obese children and adolescents with Hispanic ancestry who were living in the United States (44). The study showed that the HFF was not significantly associated with total sugar intake after controlling for demographic and anthropometric characteristics and energy intake but reported significant interactions between the HFF, PNPLA genotype and total sugar intake (44). Another cross-sectional study (rated at high ROB) examined lifestyle patterns (including diet) and metabolic variables in 38 overweight or obese children and adolescents with NAFLD diagnosed at a liver clinic and reported that “higher intakes of fructose were associated significantly with higher GGT concentrations (P=0.03)” in “best fit” multivariable models (45). Other liver enzymes such as ALT and AST were also assessed, but there were no results reported in relation to dietary fructose intakes, which indicated a high potential for a selected outcome reporting bias.

Together, we identified 6 observational studies that examined associations between dietary fructose or sucrose intake and risks of developing or the progression of NAFLD. Four studies rated at high ROB consistently reported that higher dietary fructose and sucrose intakes were associated with higher risks of developing or progression of NAFLD. However, sugar intake was not associated with indexes of liver health (HFF, ALT, and GGT) in one cross-sectional study (medium ROB) in children and one prospective cohort study (low ROB) in adults. Thus, the overall strength of evidence was rated insufficient because of high risk of biases and inconsistent study findings (Table 2).

**Intervention studies**

Twenty-one studies (in 23 publications (46–65, 67–69) that reported data on effects of free fructose on body weight (27), uric acid (28), glycemic effects (29), blood pressure (30), and blood lipids (31) for our outcomes of interest. Main rejection reasons were as follows: no exposures of interest (38 articles), no outcomes of interest (13 articles), fructose amount in the interventions cannot be quantified (8 articles), review article or letter to the editor (8 articles), liver cirrhosis patients (6 articles), animal study (1 article), alcoholic fatty liver disease (1 article), and not relevant (2 articles). **Two studies investigated both pure fructose and sucrose. All databases are available from gateway.ovid.com. CCRC, Cochrane Central Register of Controlled Trials; HFCS, high-fructose corn syrup.
TABLE 1
Characteristics of observational studies that examined associations of dietary fructose and/or sucrose consumption with risks of developing or the progression of NAFLD

<table>
<thead>
<tr>
<th>First author, pub year (ref) [country]; study design</th>
<th>Participants (ethnicity)</th>
<th>Total sample size</th>
<th>M Base characteristics</th>
<th>BMI or body weight</th>
<th>Dietary assessment methods</th>
<th>Dietary fructose and/or sucrose exposures</th>
<th>Outcomes definitions</th>
<th>Confounders adjusted</th>
<th>Funding source</th>
<th>Risk of bias</th>
</tr>
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<tbody>
<tr>
<td><strong>Adults</strong></td>
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<td>Goletzke, 2013 (66) [Australia]; prospective cohort</td>
<td>Participants from the Blue Mountains Eye Study who had provided a complete and plausible FFQ and a fasting blood specimen at follow-up. Participants who consumed &gt;20 g alcohol were excluded (predominantly white)</td>
<td>886</td>
<td>37</td>
<td>67 (NR)</td>
<td>36.8</td>
<td>FFQ</td>
<td>Sugar intake (percentage of total carbohydrate intake)</td>
<td>ALT and GGT were determined by using commercial kits performed on an automated analyzer</td>
<td>Sex, energy (use of the multivariate energy-density model), and fiber intake from fruit (g/MJ)</td>
<td>Government</td>
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<td>Ouyang, 2008 (41) [United States]; case control</td>
<td>Cases were patients with biopsy-proven NAFLD in liver clinics. Controls were patients with biopsy-proven liver disease with no identifiable cause (NR)</td>
<td>73 (49 cases; 24 controls)</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>Dietary recall/history over 3-mo period</td>
<td>Daily consumption of HFCS or sugar-containing beverages (kcal/d)</td>
<td>NAFLD case definition: liver biopsy, abnormal liver aminotransferases, and hyperechoic liver by using ultrasound imaging</td>
<td>Matching NAFLD cases with controls by age, sex, and BMI</td>
<td>Government and industry</td>
</tr>
<tr>
<td>Thuy, 2008 (42) [Germany]; case control</td>
<td>Patients undergoing liver biopsy for liver metastasis (NR)</td>
<td>18 (12 NAFLD cases; 6 controls)</td>
<td>Cases: 75; controls: 34</td>
<td>Cases: 47 (NR); controls: 55 (NR)</td>
<td>Cases: 47.8; controls: 22.5</td>
<td>Assessment by a nutritionist</td>
<td>Total fructose and sucrose intake (g/d)</td>
<td>NAFLD case definition: ultrasound and blood variables (eg, ALT, AST, and GGT)</td>
<td>None</td>
<td>NR</td>
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<tr>
<td>Volynets, 2012 (43) [Germany]; case control</td>
<td>Patients from Tubingen University Hospital, cases with NAFLD, and controls with no NAFLD (NR)</td>
<td>30 (20 NAFLD cases; 10 controls)</td>
<td>Cases: 45; controls: 30</td>
<td>Cases: 41.9 (NR); Controls: 39.6 (NR)</td>
<td>Cases: 33.1; controls: 23.1</td>
<td>FFQ</td>
<td>Total fructose and sucrose intake (g/d)</td>
<td>NAFLD case definition: ultrasonography and blood variables (eg, ALT, AST, and GGT)</td>
<td>None</td>
<td>Nonprofit</td>
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<td><strong>Children</strong></td>
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<td>Davis, 2010 (44) [United States]; cross-sectional</td>
<td>Children from community (Hispanic)</td>
<td>153</td>
<td>25</td>
<td>NR (8-18)</td>
<td>31.6 (1.95)</td>
<td>Three 24-h dietary recalls (n = 30); 3-d diet records (n = 123)</td>
<td>Total sugar intake (kcal/d)</td>
<td>Hepatic fat fraction measured by using MRI</td>
<td>Sex, age, energy, BMI, and visceral adipose tissue</td>
<td>Government and industry</td>
</tr>
<tr>
<td>Mager, 2010 (45) [Canada]; cross-sectional</td>
<td>Children from a liver clinic showing fatty liver (48% white, 28% Hispanic, 21% Asian, and 3% other)</td>
<td>38 or 35 (inconsistent reporting)</td>
<td>68</td>
<td>14.1 (5.5–19.9)</td>
<td>30.2 (1.99)</td>
<td>3-d food-intake records (2 weekdays and 1 weekend) (n = 35)</td>
<td>Total fructose intake</td>
<td>ALT, AST, and GGT from blood test after 12-h fast</td>
<td>Adiponectin and triglycerides</td>
<td>Nonprofit and industry</td>
</tr>
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</table>

1 ALT, alanine aminotransferase; AST, aspartate aminotransferase; FFQ, food-frequency questionnaire; GGT, γ-glutamyl transpeptidase; HFCS, high-fructose corn syrup; NAFLD, nonalcoholic fatty liver disease; NR, not reported; pub, publication; ref, reference.
2 All values are means.
3 All values are means; BMI z scores in parentheses.
4 OCD Fusion 5.1; Ortho Clinical.
5 Grade scale used for NAFLD diagnosis was as follows: grade 0 = steatosis, grade 1 = mild steatosis, grade 2 = moderate steatosis, and grade 3 = severe steatosis.
6 No difference in macronutrients or energy between methods.
TABLE 2  Summary of evidence from 6 observational studies that examined the associations of dietary fructose and/or sucrose consumption with risks of developing or progression of NAFLD

<table>
<thead>
<tr>
<th>Outcome of interest; population</th>
<th>Studies (total sample size)</th>
<th>Risk of bias (ref), consistency, directness, precision</th>
<th>Key findings</th>
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<tr>
<td>NAFLD (various diagnostic criteria); adults</td>
<td>Three case-control studies (81 cases; 40 hospital controls)</td>
<td>Three high risk (41–43), consistent, direct, imprecise</td>
<td>Unadjusted analyses showed significantly higher mean (± SE) daily fructose intake in NAFLD cases (n = 32) than controls (n = 16) (42, 43); NAFLD cases: 52 ± 2.5 to 58 ± 4.4 g/d (±208–232 kcal/d from dietary fructose); controls: 40 ± 3.8 to 41 ± 3.2 g/d (±160–164 kcal/d from dietary fructose)</td>
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<td>Hepatic fat fraction (MRI); Hispanic children from community</td>
<td>One cross-sectional study (n = 153)</td>
<td>One medium risk (44), NA</td>
<td>Hepatic fat fraction (%) was not significantly associated with total dietary sucrose intake (percentage of kcal/d or g/d with energy as a covariate)</td>
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<tr>
<td>Liver enzymes; children with fatty liver and older adults</td>
<td>One cross-sectional study in children (n = 38); one prospective cohort study in older adults (n = 886)</td>
<td>One high risk (45) and one low risk (66), inconsistent, indirect, precise</td>
<td>&quot;In the ‘best fit’ multivariate models (r² = 0.96, P=0.001),...omitted)...higher intakes of fructose were associated significantly with higher GGT levels (P=0.03)&quot; in children with fatty liver (45)</td>
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<td>Sugar intake (percentage of total carbohydrate intake) was not associated with ALT or GGT in multivariable concurrent change analyses from baseline to 5-y follow-up in older adults (66)</td>
</tr>
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</table>

Hypercaloric fructose compared with weight-maintenance diets (positive energy comparison)

Four studies from a research group in Switzerland compared effects of a hypercaloric fructose diet with a weight-maintenance diet on IHCLs in a total of 81 healthy, male adults (48, 54, 56, 58, 64). The earliest study assessed the effect of a 4-wk high-fructose diet (1.5 g fructose · kg body weight 

Effects of monosaccharide fructose on liver fat and other indexes of liver health in adults

Six short-term (≤4 wk) intervention studies (2 at medium, 4 at high ROB) investigated effects of a hypercaloric fructose diet (the majority delivered as fructose-sweetened drinks) on IHCLs by using proton magnetic resonance spectroscopy (a liver fat measure) (48–50, 54, 56, 58, 64). Of these publication, 3 studies also examined liver enzymes as secondary outcomes (48, 56, 58). In addition, 4 other short-term intervention studies (1 at low, 2 at medium, and 1 at high ROB) investigated effects of a hypercaloric fructose diet (delivered as fructose-sweetened drinks) on liver enzyme (AST, ALT, and/or GGT) (46, 53, 57) or hepatic DNL (55) outcomes. Furthermore, 2 other short-term intervention studies (both at medium ROB) examined the effects of an isocaloric fructose diet on bilirubin (47) or liver enzyme (52) outcomes. Neither study specified a primary outcome.

ALT, alanine aminotransferase; GGT, γ-glutamyl transpeptidase; HFCS, high-fructose corn syrup; NA, not applicable; NAFLD, nonalcoholic fatty liver disease; NR, not reported; ref, reference.

62, 63, 67, 68), and bilirubin outcomes (47, 63). Two studies investigated effects of both fructose (pure fructose or HFCS) and sucrose (46, 59). These 19 studies were in adults without NAFLD (mostly healthy, young men). In addition, 2 studies investigated effects of a fructose-reduction diet on IHCLs or liver enzymes in patients with NAFLD (69) or children with NAFLD (65). An evidence map of these 21 intervention studies by intervention-comparator pairs and outcomes of interest is shown in Figure 2. Of these publications, 13 trials were randomized controlled studies, 2 trials were nonrandomized controlled studies, and 6 trials were before-and-after studies without a concurrent control. Study characteristics are shown in Table 3.

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Six short-term (≤4 wk) intervention studies (2 at medium, 4 at high ROB) investigated effects of a hypercaloric fructose diet (the majority delivered as fructose-sweetened drinks) on IHCLs by using proton magnetic resonance spectroscopy (a liver fat measure) (48–50, 54, 56, 58, 64). Of these publication, 3 studies also examined liver enzymes as secondary outcomes (48, 56, 58). In addition, 4 other short-term intervention studies (1 at low, 2 at medium, and 1 at high ROB) investigated effects of a hypercaloric fructose diet (delivered as fructose-sweetened drinks) on liver enzyme (AST, ALT, and/or GGT) (46, 53, 57) or hepatic DNL (55) outcomes. Furthermore, 2 other short-term intervention studies (both at medium ROB) examined the effects of an isocaloric fructose diet on bilirubin (47) or liver enzyme (52) outcomes. Neither study specified a primary outcome.
showed that high-fructose diets (3.5 g · kg fat-free mass\(^{-1} \cdot \text{d}^{-1}\); 30% or 35% of energy above the energy requirement) marginally increased ALT concentrations (pooled mean difference: 4.85 U/L; 95% CI: −0.03, 9.72 U/L; \( P = 0.05 \)) compared with consumption of a weight-maintaining diet in a total of 43 healthy men and 8 women, with significant heterogeneity across studies (\( I^2 = 70\% \), \( P = 0.009 \)) (Figure 4A). Of these studies, only one RCT included a subgroup of adult women (53). With the exclusion of women, our meta-analysis reached significance with reduced heterogeneity (pooled mean difference: 6.70 U/L; 95% CI: 2.44, 10.96 U/L; \( P = 0.002 \), \( I^2 = 35\% \)). A nonrandomized controlled trial investigated the same high amount of fructose diet (3.5 g fructose · kg body weight\(^{-1} \cdot \text{d}^{-1} \) added to the weight-maintenance diet) in healthy, young adult men by comparing it with a high-fat diet (low fructose) and a high-fructose plus high-saturated fat diet (58). The study concluded that “only the FruFat diet [high fructose plus high saturated fat diet] increased ALT (by +70%)”. Furthermore, a before-and-after trial (medium ROB) showed that all liver enzyme concentrations increased after a high-fructose diet (200 g/d) for 2 wk in 74 healthy, older adult men (57): AST increased by 9 IU/L (\( P < 0.001 \)), ALT increased by 3 IU/L (\( P < 0.01 \)), and GGT increased by 13 IU/L (\( P < 0.001 \)).

For the hepatic DNL outcome, one before-and-after trial (high ROB) showed that a hypercaloric high-fructose diet (3 g/kg body weight adding to a weight-maintenance diet) significantly increased hepatic DNL by 7.8% (95% CI: 5.8%, 9.8%) in 7 healthy men (55).

Together, there was a low level of evidence that a hypercaloric fructose diet compared with a weight-maintenance diet could increase liver fat and AST concentrations in healthy men. However, there was a lack of independent repetition of study findings outside of a single research group in Switzerland and a high potential for a selected outcome reporting bias that could have led to a false-positive meta-analysis finding. There was insufficient evidence to draw a conclusion for effects on hepatic DNL on the basis of results from a single RCT.

**Hypercaloric diets high in fructose or glucose (neutral energy comparison)**

Three short-term RCTs (one crossover and 2 parallel RCTs) examined effects of fructose compared with glucose overfeeding on IHCLs (48–50, 67). The crossover RCT (medium ROB) was conducted by the Swiss research group and compared hypercaloric high-fructose with high-glucose diets (3.5 g fructose or glucose · kg body weight\(^{-1} \cdot \text{d}^{-1} \) added to a weight-maintenance diet) in 11 healthy, moderately physically active, young adult men (48). One parallel RCT (high ROB), the Tuebingen Fructose or Glucose study, was conducted in 20 healthy, German, middle-aged adult men and women (49, 50), and another parallel RCT (low ROB) was conducted in 32 centrally overweight men living in the United Kingdom (67). In all 3 studies, fructose or glucose (dissolved in water) was consumed 3 or 4 times/d with main meals. Mean IHCL concentrations were significantly increased by both fructose and glucose diets (48–50, 67). The crossover RCT (medium ROB) showed that hypercaloric high-fructose diets (3.5 g fructose or glucose · kg body weight\(^{-1} \cdot \text{d}^{-1} \)) increased ALT (by +70%) and AST (by +35%). A nonrandomized controlled trial investigated the same high amount of fructose diet (3.5 g fructose · kg body weight\(^{-1} \cdot \text{d}^{-1} \) added to the weight-maintenance diet) in healthy, young adult men by comparing it with a high-fat diet (low fructose) and a high-fructose plus high-saturated fat diet (58). The study concluded that “only the FruFat diet [high fructose plus high saturated fat diet] increased ALT (by +70%)”. Furthermore, a before-and-after trial (medium ROB) showed that all liver enzyme concentrations increased after a high-fructose diet (200 g/d) for 2 wk in 74 healthy, older adult men (57): AST increased by 9 IU/L (\( P < 0.001 \)), ALT increased by 3 IU/L (\( P < 0.01 \)), and GGT increased by 13 IU/L (\( P < 0.001 \)).

For the hepatic DNL outcome, one before-and-after trial (high ROB) showed that a hypercaloric high-fructose diet (3 g/kg body weight adding to a weight-maintenance diet) significantly increased hepatic DNL by 7.8% (95% CI: 5.8%, 9.8%) in 7 healthy men (55).

Together, there was a low level of evidence that a hypercaloric fructose diet compared with a weight-maintenance diet could increase liver fat and AST concentrations in healthy men. However, there was a lack of independent repetition of study findings outside of a single research group in Switzerland and a high potential for a selected outcome reporting bias that could have led to a false-positive meta-analysis finding. There was insufficient evidence to draw a conclusion for effects on hepatic DNL on the basis of results from a single RCT.

**Hypercaloric diets high in fructose or glucose (neutral energy comparison)**

Three short-term RCTs (one crossover and 2 parallel RCTs) examined effects of fructose compared with glucose overfeeding on IHCLs (48–50, 67). The crossover RCT (medium ROB) was conducted by the Swiss research group and compared hypercaloric high-fructose with high-glucose diets (3.5 g fructose or glucose · kg body weight\(^{-1} \cdot \text{d}^{-1} \) added to a weight-maintenance diet) in 11 healthy, moderately physically active, young adult men (48). One parallel RCT (high ROB), the Tuebingen Fructose or Glucose study, was conducted in 20 healthy, German, middle-aged adult men and women (49, 50), and another parallel RCT (low ROB) was conducted in 32 centrally overweight men living in the United Kingdom (67). In all 3 studies, fructose or glucose (dissolved in water) was consumed 3 or 4 times/d with main meals. Mean IHCL concentrations were significantly increased by both fructose and glucose diets (48–50, 67), but there was no significant difference between the 2 monosaccharides (pooled mean percentage change: −5.7%; 95% CI: −50%, 36%; \( I^2 = 0\% \)) (Figure 5).

For liver enzyme outcomes, 2 crossover RCTs (1 at low and 1 at high ROB) and 2 parallelled RCTs (both at medium ROB) that compared hypercaloric high-fructose with high-glucose diets could be meta-analyzed (46, 48, 67, 68). Our random-effects meta-analysis showed no significant difference in effects on ALT and AST [pooled mean differences (95% CIs): 2.06 U/L (−2.31, 6.42 U/L) and 1.15 U/L (−1.49, 3.78 U/L); \( I^2 = 0\% \)] between hypercaloric high-fructose and -glucose diets (doses ranged from 40 g/d to 3.5 g fructose or glucose · kg body weight\(^{-1} \cdot \text{d}^{-1} \) added to a weight-maintenance diet) (Figure 4, B and C).
## Characteristics of intervention studies that examined effects of monosaccharide fructose, sucrose, HFCS, other fructose-supplemented, or fructose-reduced diets on indexes of liver health

<table>
<thead>
<tr>
<th>First author, pub year (ref)</th>
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<th>Participants (ethnicity)</th>
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<th>Baseline age, (range)</th>
<th>BMI or body weight</th>
<th>Source of dietary fructose in interventions</th>
<th>Interventions (participants)</th>
<th>Energy balance of study design</th>
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<th>Outcomes assessed (primary or secondary endpoint)</th>
<th>Funding source</th>
<th>Risk of bias</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adults: Aeberli, 2011 (46) [Switzerland]; crossover RCT</td>
<td>Healthy M living in Zurich (NR)</td>
<td>29/24 100.0 26.3 (NR)</td>
<td>22.4 kg/m²</td>
<td>Fructose, sucrose, or glucose sweetened SSBs</td>
<td>Hypercaloric</td>
<td>3 wk (4 wk)</td>
<td>AST, ALT (secondary)</td>
<td>Nonprofit and industry</td>
<td>Low</td>
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<tr>
<td>Adults: Al-Waili, 2003 (52) [United Arab Emirates]; before-and-after trial</td>
<td>Healthy volunteers from medical staff (NR)</td>
<td>10/10 70.0 31.2 (20–45)</td>
<td>NR</td>
<td>Honey, 1.2 g/kg body weight</td>
<td>Hypercaloric</td>
<td>2 wk</td>
<td>ALT, AST, ALP (NR)</td>
<td>NR</td>
<td>Medium</td>
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<tr>
<td>Adults: Bravo, 2013 (59) [United States]; parallel RCT</td>
<td>Healthy men and women, not taking any prescription medicine or over-the-counter products for weight loss (NR)</td>
<td>80/64 56 38.7 (20–60)</td>
<td>23–35 kg/m²</td>
<td>HFCS-55 or sucrose-sweetened low-fat (1%) milk</td>
<td>Isocaloric (on the basis of lack of significant differences in total energy intake or body weight during study)</td>
<td>10 wk</td>
<td>Liver fat by unenhanced computed tomography (primary)</td>
<td>Industry</td>
<td>Medium</td>
<td></td>
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<tr>
<td>Adults: Couchepin, 2008 (53) [Switzerland]; crossover RCT</td>
<td>Healthy young M and F adults (white)</td>
<td>16/16 50.0 22.7 (NR)</td>
<td>21.8 kg/m²</td>
<td>NR</td>
<td>Hypercaloric</td>
<td>6 d (28 d)</td>
<td>ALT (NR)</td>
<td>Nonprofit</td>
<td>Medium</td>
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<tr>
<td>Adults: Fack, 2005 (55) [Switzerland]; before-and-after trial</td>
<td>Healthy, young adult M (NR)</td>
<td>7/7 100.0 NR (22–31)</td>
<td>71.5 kg</td>
<td>D-Fructose (Fluka Chemie Gmbh) in as a 20% fructose solution with 3 main meals</td>
<td>Hypercaloric</td>
<td>Fructose, 6 d (isocaloric diet given 3 d before intervention and acted as washout between interventions)</td>
<td>Hepatic de novo lipogenesis (NR)</td>
<td>Nonprofit</td>
<td>High</td>
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<tr>
<td>Adults: Johnston, 2003 (67) [United Kingdom]; parallel RCT</td>
<td>Healthy centrally overweight men, aged 18–50 y (NR)</td>
<td>32/32 100.0 34 (NR)</td>
<td>29 kg/m²</td>
<td>Fructose 25% total energy requirements compared with glucose 25% total energy requirements</td>
<td>Hypercaloric (period 1) and hypercaloric (period 2)</td>
<td>2 wk</td>
<td>IHCLs (primary); ALT, AST, GGT (secondary)</td>
<td>Nonprofit</td>
<td>Low</td>
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<th>Funding source</th>
<th>Risk of bias</th>
</tr>
</thead>
<tbody>
<tr>
<td>Koh, 1974 (60) [United States]; crossover RCT</td>
<td>Healthy F students at the University of Maryland (NR)</td>
<td>8/8 0.0 NR (18–23) 43.6–65.3 kg Uncooked fondant patties made with sucrose</td>
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<td>Fructose, 850 kcal/d compared with glucose, 850 kcal/d compared with isocaloric diet (n = 8)</td>
<td>Isocaloric</td>
<td>Sucrose and glucose, 4 wk (control consumed 1 wk before each period and 2 wk washout between sucrose and glucose)</td>
<td>ALKP, ALT, AST (NR)</td>
<td>NR</td>
<td>Medium</td>
</tr>
<tr>
<td>Koh, 1988 (47)</td>
<td>IGT adults: Controls: University and faculty staff (NR)</td>
<td>18/18 IGT: 33; control: 33 IGT: 164 lb; control: 145 lb</td>
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<td></td>
<td>Free fructose in packets, mixed with unsweetened fruit juice, milk, and water</td>
<td>Fructose, 15% total energy compared with glucose packets, 15% total energy (n = 18)</td>
<td>Hypercaloric</td>
<td>Bilirubin (NR)</td>
<td>NR</td>
<td>Medium</td>
</tr>
<tr>
<td>Le, 2006 (56) [Switzerland]; before-and-after trial</td>
<td>Healthy, moderately physically active, young adult M</td>
<td>7/7 100 24.7 (NR) 69.3 kg</td>
<td></td>
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<td></td>
<td>Pure fructose in 20% solution with the 3 main meals</td>
<td>Fructose, 3.5 g $\frac{kg}{d}$ representing an excess of 18% of the daily energy requirement (n = 7)</td>
<td>Hypercaloric</td>
<td>IHCLs (primary); ALT (secondary)</td>
<td>Nonprofit and industry</td>
<td>High</td>
</tr>
<tr>
<td>Le, 2009 (54) [Switzerland]; crossover RTC</td>
<td>Healthy, M adult offspring of type 2 diabetes patients and healthy M adults</td>
<td>24/24 100 24.7 (NR) NR</td>
<td></td>
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<td></td>
<td>Pure fructose in 20% solution</td>
<td>Fructose, 3.5 g $\frac{kg}{d}$ compared with isocaloric diet (n = 24)</td>
<td>Hypercaloric</td>
<td>IHCLs (primary)</td>
<td>Nonprofit and industry</td>
<td>High</td>
</tr>
<tr>
<td>Maaersk, 2012 (66) [Denmark]; parallel RCT</td>
<td>Healthy, nondiabetic, middle-aged adults</td>
<td>604/7 37 38.7 (20-50) 32 kg/m²</td>
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<td>Sucrose- sweetened cola (Coca Cola)</td>
<td>Sucrose 106 g/d (n = 10); compared with diet cola (n = 12) compared with semi-skimmed milk (n = 12) compared with water (n = 13)</td>
<td>Hypercaloric</td>
<td>IHCLs (primary)</td>
<td>Government, nonprofit, and industry</td>
<td>High</td>
</tr>
<tr>
<td>Ngo Sock, 2010 (48) [Switzerland]; crossover RCT</td>
<td>Healthy, moderately physically active adult M</td>
<td>11/11 100 24.6 (NR) NR</td>
<td></td>
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<td>Pure fructose in 20% solution</td>
<td>Fructose, 3.5 g $\frac{kg}{d}$ compared with glucose, 3.5 g $\frac{kg}{d}$ with weight-maintenance diet (n = 11)</td>
<td>Hypercaloric</td>
<td>IHCLs (primary); ALT, AST (secondary)</td>
<td>Nonprofit</td>
<td>Medium</td>
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<tr>
<td>Perez-Polo, 2010 (57) [Spain]; before-and-after trial</td>
<td>Healthy, older adult M (NR)</td>
<td>74 100.0 51 (NR) 28.5 kg/m²</td>
<td></td>
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<td></td>
<td>Free fructose in 10% solution</td>
<td>Fructose, 200 g/d (n = 38); fructose with allopurinol, 200 g/d (n = 38)</td>
<td>Hypercaloric</td>
<td>ALT, AST, GGT (secondary)</td>
<td>Nonprofit</td>
<td>Medium</td>
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<tr>
<td>Porikos, 1983 (62) [United States]; before-and-after trial</td>
<td>Middle-aged adult M (NR)</td>
<td>21</td>
<td>100.0</td>
<td>NR (24-45)</td>
<td>NR</td>
<td>Snacks and food with sucrose</td>
<td>Sucrose, 25–30% kcal (n = 21)</td>
<td>Hypercaloric</td>
<td>30 d</td>
<td>ALT, AST (primary)</td>
<td>Government and industry</td>
</tr>
<tr>
<td>Purkins, 2004 (63) [UK]; crossover RCT</td>
<td>Healthy adult M (NR)</td>
<td>12/12</td>
<td>100.0</td>
<td>NR (20-41)</td>
<td>NR</td>
<td>Readily available sucrose-containing food</td>
<td>High-carbohydrate diet (sucrose, 1408 kcal/d; 4500 kcal/d), compared with high-fat diet (sucrose 180 kcal/d; 4500 kcal/d) compared with standard diet (1900 kcal/d) (n = 12)</td>
<td>Hypercaloric</td>
<td>8 d (14-d washout)</td>
<td>ALP, ALT, AST, GGT, bilirubin (primary)</td>
<td>Industry</td>
</tr>
<tr>
<td>Silbernagel, 2011 (50); Silbernagel, 2012 (49) [Germany]; parallel RCT</td>
<td>Healthy, middle-aged adults</td>
<td>25/20</td>
<td>60</td>
<td>30.5 (20–30)</td>
<td>25.9 kg/m²</td>
<td>Pure fructose powder dissolved in water</td>
<td>Fructose, 150 g/d (n = 10) compared with glucose, 180 g/d (n = 10)</td>
<td>Hypercaloric</td>
<td>4 wk</td>
<td>IHCLs (primary)</td>
<td>Nonprofit</td>
</tr>
<tr>
<td>Sobrecases, 2010 (58) [Switzerland]; non-RCT</td>
<td>Healthy adult M</td>
<td>30/30</td>
<td>100</td>
<td>23.9 (NR)</td>
<td>22.6 kg/m²</td>
<td>NR</td>
<td>Fructose: 3.5 g/kg (n = 12) compared with high fat, 30% total energy as saturated and polyunsaturated fats (n = 10), compared with fructose plus high fat (n = 8)</td>
<td>Hypercaloric</td>
<td>Fructose: 7 d, high fat and fructose and high fat: 4 d</td>
<td>IHCLs (primary), ALT (secondary)</td>
<td>Nonprofit</td>
</tr>
<tr>
<td>Stanhope, 2009 (51); Cox, 2012 (68) [United States]; parallel RCT</td>
<td>Healthy, older adults (NR)</td>
<td>First 23 enrolled people/18 (DNL outcome); 30/32 (liver enzyme outcomes)</td>
<td>50.0</td>
<td>53.7 (NR)</td>
<td>29.3 kg/m²</td>
<td>Free fructose added to unsweetened beverages (Kool-Aid; Kraft)</td>
<td>Fructose (&lt;25% of daily energy) (n = 10) compared with glucose SSB (&lt;25% of daily energy) (n = 8)</td>
<td>Hypercaloric</td>
<td>10 wk</td>
<td>Hepatic de novo lipogenesis, ALT, AST, GGT (NR)</td>
<td>Government</td>
</tr>
<tr>
<td>Theytaz, 2012 (64) [Switzerland]; crossover RCT</td>
<td>Healthy, nonobese, nonsmokers, and sedentary M</td>
<td>99</td>
<td>100</td>
<td>23.3 (NR)</td>
<td>22.6 kg/m²</td>
<td>Pure fructose provided as drinks 5 times/d</td>
<td>Fructose, 3 g/kg, + essential amino acid (&lt;38% of daily energy) compared with Fructose, 3 g/kg plus placebo (&lt;30% of daily energy) compared with weight-maintenance diet (n = 9)</td>
<td>Hypercaloric</td>
<td>6 d (4–10-wk washout)</td>
<td>IHCLs (primary)</td>
<td>Industry</td>
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TABLE 3  (Continued)

<table>
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<tr>
<th>First author, pub year (ref) [country]; study design</th>
<th>Participants (ethnicity)</th>
<th>Participants enrolled/analyzed</th>
<th>M</th>
<th>Baseline age, (range)²</th>
<th>BMI or body weight¹</th>
<th>Source of dietary fructose in interventions</th>
<th>Interventions (participants)</th>
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</thead>
<tbody>
<tr>
<td>Volynets, 2012 (69) [Germany]; before-and-after trial</td>
<td>Patients with NAFLD, diagnosed by ultrasound and blood variables (NR)</td>
<td>15/10</td>
<td>40</td>
<td>45.5 (34.5–51.5)</td>
<td>31.1</td>
<td>Reduction in the consumption of fructose-rich foods (eg, to avoid sweets, lemonades, fruit juices) and to prefer foods with a lower content of fructose</td>
<td>Nutritionists advised patients to reduce their daily fructose intake by 50%</td>
<td>Hypocaloric</td>
<td>6 mo</td>
<td>IHCLs (primary); ALT, AST, GGT (secondary)</td>
<td>Nonprofit</td>
<td>High</td>
</tr>
<tr>
<td>Vos, 2009 (65) [United States]; parallel RCT</td>
<td>Children with NAFLD (NR)⁶</td>
<td>10</td>
<td>NR</td>
<td>13.0 (2.1)</td>
<td>Elimination of sugar-containing beverages, fruit juice, and HFCS</td>
<td>Low fructose (&lt;20 g/d fructose intake) (n = 6) compared with low fat (&lt;15 g/d fructose intake) (n = 4)⁷</td>
<td>Hypocaloric</td>
<td>6 mo</td>
<td>ALT, AST (secondary)</td>
<td>Nonprofit and government</td>
<td>High</td>
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¹ ALT, alanine aminotransferase; ALKP, alkaline phosphatase; ALP, alkaline phosphatase; AST, aspartate aminotransferase; DNL, de novo lipogenesis; GGT, γ-glutamyl transpeptidase; HFCS, high-fructose corn syrup; HFCS-55, high-fructose corn syrup (55% fructose and 45% glucose); IGT, impaired glucose intolerant; IHCL, intrahepatocellular lipid; NR, not reported; pub, publication; RCT, randomized controlled trial; ref, reference; SSB, sugar sweetened beverage.

² All values are means; ranges in parentheses.

³ All values are means or ranges; BMI z score in parentheses.

⁴ Study design included fish oil compared with fish oil and fructose compared with fructose. Fish-oil interventions were not included in the analysis.

⁵ A nonrandomized crossover trial. Ranges of fructose intake reported were as follows: fructose and glucose = for IGT, 45–83 g/d; for control, 45–112 g/d.

⁶ Fifteen normal-weight and 6 overweight subjects.

⁷ Ten-week duration included an 8-wk hypercaloric diet and 2-wk isocaloric diet.

⁸ Seven children (70%) with a confirmatory liver biopsy that showed nonalcoholic steatohepatitis.

⁹ Low fructose denoted the elimination of sugar-containing beverages, fruit juice, and food items that were high in high-fructose corn syrup. Low fat according to the American Heart Association recommendation.
For the hepatic DNL outcome, one 10-wk RCT (high ROB) showed that postprandial DNL, but not fasting hepatic DNL, was significantly increased by 48% (95% CI: 28.7%, 104%) when hypercaloric high-fructose and high-glucose diets (+25% of daily energy) were compared in 18 healthy older adults (51). Together, there was a low level of evidence that hypercaloric fructose and glucose diets have similar effects on liver fat and liver enzymes in healthy adults. There was insufficient evidence to draw a conclusion for effects on hepatic DNL on the basis of results from a single RCT. Note that the hypercaloric nature of the diets may have obscured any differences between the 2 monosaccharides were any to exist.

Effects of isocaloric fructose intake in adults

Three short-term intervention studies (1 at low and 2 at medium ROB) investigated effects of isocaloric fructose intake (47, 52, 67). Studies could not be synthesized together because they investigated different comparisons. One 2-wk parallel RCT (low ROB) examined effects of an isocaloric diet with 25% of the daily energy requirement from fructose or glucose on IHCLs and liver enzyme outcomes (ALT, AST, and GGT) in 32 centrally overweight men who were living in the United Kingdom (67). The 2 isocaloric monosaccharide diets did not alter IHCLs (+0.11% ± 2.1%) but reduced liver enzyme concentrations slightly. However, the 2 isocaloric fructose and glucose diets did not differ in any hepatic outcome measure. One 4-wk controlled trial compared effects of an energy-balanced diet with 15% of total calories (45–83 g/d) from fructose or glucose (both sugars were given in packets and mixed with unsweetened fruit juice, milk, or water) in 9 adults with impaired glucose intolerance and 9 healthy adults (47). The study showed that total bilirubin concentrations increased significantly in adults with impaired glucose intolerance (+0.45 mg/dL) but not fructose consumption (−0.9 mg/dL). Total bilirubin concentrations did not change in healthy adults with normal glucose-tolerance. Another 2-wk before-and-after study (medium ROB) showed that a controlled regular diet supplemented with the daily consumption of 1.2 g honey/kg body weight dissolved in 250 mL water (containing 38 g% fructose, 30 g% glucose, and some vitamins and minerals) decreased concentrations of AST by 22% and ALT by 18% in 10 healthy volunteers from the medical staff (52).

Together, there was insufficient evidence to draw a conclusion on effects of isocaloric fructose intake (from pure fructose or honey) on indexes of liver health on the basis of results from a single study per exposure-comparison pair.

Effects of HFCS compared with sucrose on liver fat in adults

A 10-wk RCT (medium ROB) compared effects of a HFCS-sweetened beverage and sucrose-sweetened low-fat (1%) milk on liver fat deposition measured by using computed tomography (59). Eighty healthy adult men and women were challenged with the 3 amounts of HFCS-55 (ie, 55% fructose and 45% glucose) or sucrose-sweetened low-fat milk at 8%, 18%, or 30% of the energy requirement for weight maintenance. These amounts are equivalent to the 25th, 50th, and 90th percentiles, respectively, for fructose consumption in the general population. All participants were
instructed to substitute test beverages for food or other beverages usually consumed. No additional instruction was given. Non-compliance (defined as the cumulative consumption of test beverages 80% or 5 consecutive days without the consumption of a single serving of the test beverage) led to automatic withdrawal from the study. Analyses were done in 64 participants who completed the study (dropout: 20%). During the 10-wk intervention period, energy intake increased across the entire cohort and was driven by an increase in intake of total sugar (mean: 97 compared with 216 g/d; \( P < 0.001 \)), which resulted in a mean weight gain of 0.8 kg independent of the diet group. Despite the weight gain, there were no significant changes in liver fat (mean: 13.32% compared with 13.21%) regardless of the type or amount of beverage.

No other studies investigated effects of HFCS. Thus, there was insufficient evidence to draw a conclusion for effects of HFCS on liver fat on the basis of results from a single RCT.

Effects of sucrose on liver fat and other indexes of liver health in adults

For the liver fat outcome, one 6-mo parallel RCT (high ROB) evaluated effects of 4 commercially available beverages [sucrose-sweetened regular cola (Coca Cola), aspartame-sweetened diet cola (Coca Cola), semi-skimmed milk (Arla Foods), and still mineral water (AQUA D’OR)] on ectopic fat accumulation in the liver in 60 healthy, normoglycemic individuals who were living in Denmark (61). IHCL was determined by using proton magnetic resonance spectroscopy. Study participants were provided with 1 L test drink/d and allowed to drink water, tea, coffee, and their usual amounts of alcohol. The 1 L sucrose-sweetened regular cola (106 g sucrose/d = 53 g bound fructose/d), semi-skimmed milk, aspartame-sweetened diet cola, and mineral water provided 430, 451, 4, and 0 kcal/d, respectively. The dropout or termination (all women) was 22%. Analyses were performed in 47 individuals who completed the study and were adjusted for sex. At the end of the intervention period, IHCL was significantly increased in the sucrose-sweetened regular cola group (\( n = 10; +132\% \)) but not in the 3 other beverage groups. IHCL was similar in the semi-skimmed milk, diet cola, and water groups (61). There were no significant differences in total energy intake or body weight in the 4 groups during the study, which suggested energy compensation in response to increased intake of the 2 energy-containing beverages regular cola and semi-skimmed milk.
Three crossover RCTs (46, 60, 63) and one before-and-after study (62) examined effects of hypercaloric diets high in sucrose on liver enzymes or other indexes of liver health outcomes. These 4 studies could not be synthesized together because of different comparisons. One crossover RCT (low ROB) reported no significant differences in AST and ALT between hypercaloric high-sucrose (40 or 80 g/d) and high-glucose diets in 24 healthy men (46). Another crossover RCT (at medium ROB) also showed no significant differences in AST and ALT after energy-balance diets with high sucrose (850 kcal/d) or high glucose in 8 healthy, female university students (60). On the contrary, a crossover RCT (high ROB) showed that all liver function test indexes (alkaline phosphatase, ALT, AST, GGT, and bilirubin) were significantly raised after a hypercaloric high-sucrose (double energy requirement with 32% of energy from sucrose) diet compared with a standard energy-balanced diet in 12 healthy men (63). The before-and-after trial (high ROB) also showed that a hypercaloric sucrose-containing food-supplemented diet (25–30% kcal) significantly increased ALT and AST concentrations (62).

Together, there was insufficient evidence to draw a conclusion for effects of sucrose on liver fat and other indexes of liver health outcomes in adults on the basis of results from a single study for each exposure-comparison pair within each outcome of interest as well as high risks for biases.

**Effects of hypocaloric fructose diets in NAFLD patients**

One parallel RCT (high ROB) in 10 obese children with NAFLD (65) and one before-and-after trial (high ROB) in 10 overweight adults with NAFLD (69) examined the effects of a fructose-reduction diet on IHCLs or liver enzymes. The RCT in obese children evaluated diet education of a low-fructose (eliminating sugar-containing beverages, fruit juice, and food items with HFCS) and compared it with diet education of a low-fat diet (American Heart Association recommendations), and results showed no significant changes in ALT and AST concentrations in either group after 6 mo (65). However, children’s BMI z scores did not change significantly during the study. The before-and-after trial in overweight adults assessed the effect of a dietary intervention to reduce fructose intake by 50% on IHCLs and liver enzyme concentrations (69). Patients had reduced their fructose intakes by an average of 61% along with significant reductions in average daily intake of total calories (~36%), total fat (~31%), and saturated fat (~24%) after the 6-mo intervention. After 6 mo, the mean IHCL content was significantly reduced (~36%), and AST and ALT concentrations were within the normal range, whereas GGT concentrations were lower in 7 of 10 patients. Patients’ body weight and BMI were significantly lowered after 6 mo as result of the hypocaloric dietary intervention.

Together, there was insufficient evidence to draw a conclusion for effects of a hypocaloric fructose diet on the progression of NAFLD in patients with NAFLD because of high risks for biases, although both studies did not find that a hypocaloric fructose diet had a significant effect on ALT and AST concentrations.

**DISCUSSION**

We found scarce, poor-quality, and heterogeneous data (different exposure comparisons) on effects of different amounts and
forms of dietary fructose on risks of developing or the progression of NAFLD and on indexes of liver health in humans (Figure 2). For observational studies, the overall strength of evidence for associations of dietary fructose or sucrose consumption with risks of developing or the progression of NAFLD was rated insufficient because of high risk of biases and inconsistent study findings. Note that the causality between dietary fructose and NAFLD could not be assessed from retrospective case-control and cross-sectional studies. Of the 21 intervention studies identified, 12 studies (57%) investigated effects of a hypercaloric fructose diet (supplemented by pure fructose) in almost exclusively healthy, young, male adults. Of these studies, 8 studies (67%) were conducted by the same group of investigators. Few intervention studies have directly distinguished between effects of increases in energy intake or body weight from high intakes of monosaccharides and disaccharides, particularly of fructose, per se on liver fat or NAFLD. Within the context of current intakes of fructose in the United States, there are no studies, to our knowledge, that have compared liquid and solid forms of fructose on liver fat or NAFLD. Only one study directly compared the isocaloric replacement of fructose and glucose (25% kcal) on liver fat and liver enzyme outcomes and showed the 2 monosaccharides did not differ in any hepatic outcome measure (67). Of studies for which some data were available, common limitations included small sample sizes, short intervention periods, a lack of energy intake or body weight control, the use of pure fructose rather than sucrose or HFCS, and fructose loads that exceed current intakes. With these caveats in mind, we concluded that there was a low level of evidence that a hypercaloric fructose diet increases liver fat and AST concentrations in healthy men compared with a weight-maintenance diet in healthy men. However, it should be stressed that liver fat concentrations observed in healthy men at baseline and after fructose overfeeding were still far below concentrations observed in patients with NAFLD (70). Furthermore, the comparison of the substitution of glucose to fructose in hypercaloric diets may not be directly representative of that encountered in free-living individuals who consume fructose and glucose not only as monosaccharides but also has disaccharides and dextrans. In addition, there was a low level of evidence that hypercaloric fructose and glucose diets have similar effects on liver fat and liver enzymes in healthy adults. Note that the elevation of liver fat or liver enzyme concentrations does not automatically imply the presence of liver disease; the concentrations of liver enzymes do not correlate well with the extent of liver damage nor prognosis (71, 72). Our conclusions are consistent with a recently published systematic review and meta-analysis that examined the effect of fructose on markers of NAFLD in controlled trials although the authors pooled studies that reported IHCL outcomes by using a different metric (standardized mean difference) (73). The consistent findings further support the strengths of our approaches to conducting both qualitative and quantitative syntheses of data in light of the significant clinical and statistical heterogeneity observed in available data as well as the poor quality of the data.

With the potential to progressing to advance liver disorders, NAFLD has been recognized as a major public health concern (74, 75). Diet is considered to play a central role in the development of NAFLD through its effects on hormones, transcription factors, and lipid metabolism (5). However, it difficult to isolate causes to a single dietary cause because of the high prevalence of NAFLD in obese individuals and patients with diabetes (76–79). Furthermore, there is currently no consensus for a noninvasive reference standard for the diagnosis of NAFLD. Limitations in invasively assessing liver fat have resulted in little data on which to base diet recommendations, and data available are, for the most part, inconclusive (80). Currently, there are no specific guidelines for the prevention or treatment of NAFLD or NASH with diet beyond weight loss, which is an independent risk factor for NAFLD (81–83).

This systematic review identified several important research gaps. First, a liver biopsy is the gold standard for diagnosing NAFLD but is unlikely to be used in large population-based studies. Without the use of the same definition of a reference standard, conducting more studies on NAFLD is unlikely to add meaningfully to the body of evidence. Therefore, the establishment of a noninvasive reference standard for NAFLD diagnosis is urgently needed to advance our knowledge in this research area. Second, well-controlled studies are needed to examine factors that may modify effects of fructose or sucrose consumption on NAFLD. For example, a genetic variation may modify effects of fructose or sugar intake on liver fat (44, 84) and determine whether certain subpopulations may be more vulnerable to the development of NAFLD. Last, future studies need to have sufficient statistical power to test hypotheses with prospective observational study designs in the general population or use experimental conditions that approximated current intakes and chemical forms (pure fructose does not exist in a typical diet), so that results can inform dietary intake recommendations for disease prevention.

Our systematic review had several strengths. Unlike other published systematic reviews on fructose and a variety of health outcomes (27–31), we evaluated all study designs and all sources and forms of fructose intake in relation to liver health with a goal to reach conclusions that are more applicable to general population. We also tailored risk-of-bias tools to include nutrition-specific quality items, which are important for evaluating the quality of nutrition research (85). Our study also had several limitations, which were primarily inherited from poor-quality and heterogeneous studies included in this systematic review. In addition, most of the data were only available in healthy men, which limited the generalizability of our findings. The dearth of data in women could not answer the question of whether sex is an effect modifier for associations between dietary fructose consumption and liver health. Finally, we showed that a selected outcome reporting bias (86) was likely in many studies, which could have led to an overestimation of effects.

In conclusion, on the basis of indirect comparisons across study findings, the apparent association between indexes of liver health (ie, liver fat, hepatic DNL, ALT, AST, and GGT) and fructose or sucrose intake appear to be confounded by excessive energy intake. Current evidence does not allow us to discern the intertwined associations between excess body weight, monosaccharide fructose or sucrose intake, and NAFLD. Therefore, we concluded that, overall, the available evidence is not sufficiently robust to draw conclusions regarding effects of fructose, HFCS, or sucrose consumption on NAFLD.

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