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Understanding susceptibility and targeting treatment in non-alcoholic fatty liver disease in children; moving the fulcrum

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Non-alcoholic fatty liver disease (NAFLD) is the most common cause of paediatric liver disease, affecting 10 % of school-aged children and 44–70 % of obese children and young people (CYP) in the western world. Encompassing a spectrum from simple steatosis to steatohepatitis and progressive fibrosis, the disease is rapidly becoming the most common indication for liver transplantation. The molecular pathogenesis of NAFLD remains only partially understood. Development and progression of NAFLD is influenced by genetic and nutritional factors, insulin resistance, oxidative stress, gut microbiome, bile acid metabolism and lipid/glucose handling and is closely associated with overweight and obesity. Lifestyle change is the only proven effective treatment for paediatric NAFLD, however this is difficult to achieve in many. Given that moderate or severe fibrosis is already present in 30–50 % of children with NAFLD at the time of presentation, progression in CYP may be more rapid, though adequate outcome data do not yet exist in this cohort. CYP with NAFLD are an excellent population in which to study underlying mechanisms and interventions to correct disease progression as they are largely unaffected by other environmental influences such as alcohol and may represent the more severe end of the spectrum in terms of early onset. Undoubtedly genetic and epigenetic mechanisms determine a large proportion of susceptibility to the disease and potentially, identification of individuals at risk may allow for targeted therapy. This review with give a clinical perspective of paediatric NAFLD focused on identifying those at risk of progressive disease and what to consider in attempting to modify risk.

Non-alcoholic fatty liver disease: Paediatric: Susceptibility: Mental health

It is now well recognised that non-alcoholic fatty liver disease (NAFLD) in adults is rapidly becoming the most common indication for liver transplantation[1]. The recent Lancet series in liver disease drew attention to the fact that mortality from liver disease (including NAFLD, alcohol and hepatitis C) continues to rise and in the UK is now at 500 % of that in the 1970s, in contrast to a decline in the standardised mortality rate for cancer, CVD and respiratory disease[2]. The number of admissions to hospital with NAFLD as the primary indication is rising dramatically and accompanies exponential increase in the healthcare burden of disease[3].

NAFLD is a condition characterised by liver steatosis with or without inflammation and fibrosis, most often in the setting of overweight or obesity[4,5]. Although typical features are seen on liver biopsy, diagnosis requires the exclusion of all other causes of liver disease including alcohol, other toxins and liver-based metabolic disease[6]. There is a real risk, although probably in minority of patients with NAFLD, of progression to end-stage liver disease and/or hepatocellular carcinoma[7]. The prevalence of the condition means that even though only a percentage of those affected will progress to end stage disease, this is still a large number and a considerable

Abbreviations: CYP, children and young people; Hh, hedgehog; HFD, high-fat diet; NAFLD, non-alcoholic fatty liver disease; NASH, non-alcoholic steatohepatitis.

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burden on an already stretched transplant service. In other words, fat in the liver remains a marker of cardiovascular risk rather than liver disease *per se*, and there is not necessarily a progression to significant fibrosis[8,9]. Previously it was thought that only those with significant inflammation and ballooning in the liver biopsy (non-alcoholic steatohepatitis (NASH)) were those who were at risk of fibrosis progression. Now it is understood that even those without NASH at time of biopsy are also at risk of progressive disease. The most relevant and important marker of prognosis is fibrosis stage, even in the absence of inflammation[7].

The rise in prominence of NAFLD, the liver manifestation of the metabolic syndrome, is not surprising given the association with the prevalence of obesity[10]. BMI does not measure the entire problem however, with reports of patients with lean NAFLD, although there is still often an association with visceral adiposity, insulin resistance and poor metabolic health[11].

The paediatric population have seen a parallel rise in prevalence of NAFLD, again likely related to the rise in obesity[3]. There is an argument that early onset disease is a more severe phenotype in that it presents early in life, often with significant fibrosis at time of diagnosis, following only a relatively short time of exposure to the implicated risk factors and usually without exposure to alcohol and other cofactors[12]. NAFLD in children and young people (CYP) may therefore serve as an excellent pathophysiological model of this heterogeneous condition and allow insights into susceptibilities and triggers. This review will focus on the differences in the adult condition, the challenges and opportunities that are specific to paediatric NAFLD. Thankfully, CYP present with a potentially modifiable condition. Clinical experience confirms that fibrosis up to and including the point of cirrhosis is often reversible once the perpetuating stimulus is removed[13] and thus changing habits during this time of life can lead to a lifetime of diminished risk.

Prevalence and presentation

Variation in reports of prevalence of NAFLD is partly due to the different methods of detection[9,14,15]. Although the gold standard for diagnosis is liver biopsy, this is not practical in population studies. One of the most veritable studies reporting prevalence in the general paediatric population is an autopsy study of CYP who died of accidental injury[16]. In this study, Schwimmer *et al*. reported a prevalence of NAFLD in 9·6 % of this cohort, with NASH (steatohepatitis) present in 3 %[16]. Other population studies in children vary in prevalence from 1·7 to 42·5 %[14]. This variation is due to the different methods used to diagnose the condition and the different population studies both in terms of ethnicity and risk factors. In a study of morbidly obese children undergoing bariatric surgery, NAFLD was identified in biopsy in 83 %[17] in contrast to a longitudinal study of healthy children from the UK midlands using ultrasound, in which the prevalence was only 2·5 %[18].

Table 1. Differential diagnosis of fatty liver in CYP

Clinical condition	Diagnostic clues
Wilson disease	Low ceruloplasmin, high urinary or tissue copper, mutational analysis
Alpha 1 antitrypsin deficiency	Phenotype or genotype
Drugs – steroids, amiodarone, alcohol, methotrexate, MDMA (ecstasy), L-asparaginase, vitamin E, valproate, tamoxifen, antiretrovirals	History
Cystic fibrosis associated liver disease	History/sweat test or mutational analysis
Malnutrition	History
Coeliac disease	Tissue transglutaminase, jejunal biopsy
Hepatitis C	HCV antibody status
Parenteral nutrition associated liver disease	History
Mitochondrial disease/fatty acid oxidase deficiency	Lactate, acylcarnitines, respiratory chain enzymes, mutational analysis
Metabolic disease: lysosomal acid lipase deficiency (cholesterol ester storage disease)	White cell enzymes, mutational analysis
Galactosaemia	Gal-1-PUT
Fructosaemia	Enzymology
Glycogen storage disease	White cell enzymes, mutational analysis
Peroxisomal disorders	Very long chain fatty acids, mutational analysis
Mauriac syndrome	History of type-1 diabetes
Hypobetalipoproteinaemia/ abetalipoproteinaemia	Low lipid levels, reduced/ absent ApoB/mutational analysis
Lipodystrophies	Mutational analysis
Shwachman syndrome	Pancreatic insufficiency/ mutational analysis

MDMA, 3,4-methyl endoxy metamphetamine; HCV, hepatitis C virus; Gal-1-PUT, galactose-1-phosphate uridyl transferase.

CYP often present with an incidental finding of elevated transaminases and an echogenic liver on ultrasound. CYP with NAFLD are likely to be overweight or obese but this is not universal[11]. Neither bloods nor imaging will differentiate those with significant fibrosis from those without[19] and a liver biopsy is still required in some. Biopsy is not practical in most cases however, given its invasiveness, the need for sedation and the inherent risks involved. Both the European and the North American Societies for Paediatric Gastroenterology, Hepatology and Nutrition recommend biopsy in children in cases of clinically suspected advanced liver disease, in those in whom another disease is a possibility, before pharmacological treatment, and as part of a structured intervention protocol or clinical research trial[20,21].

All other possible causes of liver disease in children should be ruled out including liver-based metabolic disease[6]. A family history of metabolic syndrome should be sought in addition to medication use and other risk factors. A list of differential diagnoses is given in Table 1.

As liver biopsy is not always possible or felt appropriate, non-invasive methods are often used to determine

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the severity of disease in terms of fibrosis. Likewise, non-invasive methods of longitudinal monitoring are used as repeated biopsy is only rarely undertaken[22]. Although there is no consensus in terms of appropriate non-invasive measures, transient elastography is an imaging technique which is reliable in determining no fibrosis from significant fibrosis/cirrhosis[23,24]. MRI protocols can differentiate different degrees of steatosis and magnetic resonance elastography is a promising tool for fibrosis staging, however the cost is often preclusive[25]. There are blood-marker algorithms available which have been derived from adult populations, including the NAFLD fibrosis index and Fib4[9,26], although none have been adequately validated in children. Algorithms which include age and markers of collagen turnover influenced by growth such as procollagen III peptide may not accurately reflect fibrosis in this population and rederivation of many of the available algorithms is needed. The paediatric NAFLD fibrosis index was developed using a discovery cohort of children and may be a useful tool, however external validation is required[27].

Susceptibility to developing non-alcoholic fatty liver disease in childhood

Genetics

Genetic variation as a risk factor is well established in adult populations with NAFLD. This is borne out from both genome wide association studies and candidate gene studies[28]. A smaller body of evidence exists in children with the condition, arguably a population which is more likely to demonstrate the effects of genetic variation[29].

NAFLD is clearly not a monogenetic disorder but variants in certain genes, namely *PNPLA3, TM6SF2* and *APOB* have been found consistently to convey a susceptibility to NAFLD both in adults and in children[30–35]. A single paediatric genome wide association study in a cohort of Hispanic boys has shown novel gene effects on histology distinct from those previously recognised in adult cohorts[36]. Most importantly, the effect of these variants is seen most clearly in the setting of increased BMI[37]. The combination of genetic variants may also be of importance. In a study of 450 children in whom hepatic fat was measured by using MRI hepatic fat fraction, the combination of variants: *PNPLA3* rs738409, *TM6SF2* rs58542926, *GCKR* rs1260326 and *MBOAT7* rs626283 explained 19 % of intrahepatic fat content variance[38]. The effect of multiple loci on the development of steatosis as measured by intrahepatic fat content% is amplified in the presence of overweight and obesity. This variation is in genes involved in different pathways including lipid droplet modelling, lipogenesis, oxidative stress, immune system activation and fibrogenesis[39] Understanding the genetic variation contributing to disease in an individual may allow more targeted prevention and reversal of disease.

The importance of the antenatal environment and early infant nutrition

The importance of the antenatal environment on metabolic programming has long been recognised[40]. It is known that NAFLD is part of the consequence of programming, although not understood how early in life this may manifest nor how reversible is the effect[41]. Animal models reflect that consequences of *in utero* exposure on liver metabolism[42]. The 'priming' of a child's liver, even before birth, to injury, with lipid accumulation, increased oxidative stress, apoptosis and innate immune dysfunction may play a role in the development of NAFLD in these children[43].

Rodent models have demonstrated the effect on the liver of high-fat diet (HFD) in dams and the development of fatty liver in the offspring. Methionine choline deficient diet in pups has a similar yet not as dramatic an effect[44]. Not surprisingly HFD in dams and methionine choline deficient diet in pups leads to a cumulative effect on the liver of the offspring, with both DNA methylation alteration and a decrease in diversity gut microbiome showing the effect of the maternal HFD and possibly mediating the effect on liver histology[44]. In a study of differences in outcomes of male and female offspring of obese rats, Lomas-Soria *et al.* found a more significant effect on liver in male offspring raising some important questions about differences between the sexes in terms of NAFLD susceptibility mediated by early life exposure[45].

Mouralidarane *et al.* have clearly demonstrated *in utero* HFD exposure results in lipid accumulation in addition to elevated levels of oxidative stress and impairment of innate immunity in a mouse model[43]. In this study, offspring mice were followed to 12 months following exposure to maternal obesity and a post-weaning HFD. Both factors were independent risk factors for steatosis and at 12 months for steatohepatitis and fibrosis. There was a significant increase in liver injury in those exposed to both maternal obesity and a high fat post-weaning diet. Increased mRNA expression of inflammatory cytokines and fibrogenic enzymes were also found[43].

Taken together, this 'priming' of the liver, which may then be exposed to years of excess nutrition and sedentary behaviour can result in a worse phenotype and/or more accelerated disease than would otherwise have been the case. Interestingly, a macaque model of HFD-fed macaques who were fed with a normal chow diet prior to breeding and during pregnancy produced offspring with substantially less steatosis than those who remained on the HFD[46].

Human studies also reflect the importance of early life exposure. In a study of still births to mothers with gestational diabetes, pathological findings in post-mortem infants demonstrated 78·8 % steatosis in infants of mothers with gestational diabetes *v.* 16 % in infants of mothers who were not diabetic[47]. In live-born infants, Brumbagh *et al.* investigated intrahepatic lipid content using MRI and found that this was higher in infants born to women who were obese and those with type-2 diabetes than in those born to normal weight women[48]. As deposition of adipose tissue does not occur until the 3rd trimester, there is instead hepatic storage of the excess substrate that the fetus is exposed to during pregnancy in addition to *in utero de novo* lipogenesis in response to a high transplacental glucose supply[40].

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As 25–30 % of mothers are now obese at the time of conception, *in utero* exposure is an important epidemiological phenomenon[49].

Birth weight is also relevant in the later development of NAFLD in that the disease associates with both low and high birth weight in a large cohort[50]. Again, the implication is that early exposure may mediate priming of the liver at a later stage of development suggesting that antenatal conditions may influence later presentation. The role of maternal obesity, method of delivery and breast-feeding all have a recognised effect on the infant gut microbiome which in turn is associated with later risk of obesity[41]. Infants with a decreased microbiome diversity at the age of 6 months were found to have a greater risk of obesity aged 7 years[51]. Breast milk promotes the colonisation of the intestinal microbiota transferring a low biomass of microbiota to the infant gut and providing oligosaccharides which act as prebiotics. Immune tolerance is promoted by the early pioneering gut microbiota which are decreased in number in offspring of obese women[41].

As yet there is no evidence for a specific diet in pregnancy that will increase or decrease later risk of NAFLD in offspring. Breast-feeding appears to protect against NAFLD, although studies are not conclusive[52].

Nutritional intake in childhood, growth and development

Body growth is unique to the paediatric population, necessitating higher body weight to energy requirements, compared to adults. Liver in childhood has lower probability of exposure to toxins such as alcohol and other environmental toxins. The complex interaction between nutritional toxins (saturated fats and sugar) with the liver cells of the maturing liver is less well studied however. Children and teenagers are the highest consumers of fructose[53,54], with emerging evidence that this may be implicated in the development and severity of NAFLD possibly through increasing intestinal permeability and translocation of endotoxin[55,56].

Several studies of fructose consumption and association with NAFLD severity in children have been reported[55,57]. A UK study in children with NAFLD failed to show a distinct difference in qualitative dietary intakes in children with NAFLD *v.* obese controls without NAFLD[58], although undoubtedly it is the underlying susceptibility coupled with the trigger that mediates the injurious effects. Interestingly the effects of fructose on endotoxaemia which, as earlier may be an important trigger in hepatic inflammation, were seen more prominently following fructose bolus in children with NAFLD *v.* those without NAFLD. Children were given a fructose sweetened beverage with each meal in a 24 h cross over study. In another part to this study, children with NAFLD were subjected to a fructose drink *v.* a glucose sweetened drink with meals for a 4-week period, with the fructose group demonstrating higher endotoxaemia[56]. In a study by the same group exposing children with NAFLD (*n* 9) to fructose or glucose beverage in a cross over study compared to matched controls without NAFLD (*n* 10), the TAG incremental area under the curve was higher in the fructose exposed groups and to a greater extent in those with NAFLD ($P = 0{\cdot}019$)[59].

The association of NAFLD and insulin resistance is clearly established. Periods of maximum insulin resistance during the lifetime include pregnancy and the pre-adolescent, particularly in boys. It may be the case that during these periods of maximal insulin resistance, fatty liver develops in susceptible individuals[39].

Histology and differences in paediatric non-alcoholic fatty liver disease

The NASH Clinical Research Network in the USA convened a group of expert hepatopathologists to develop a scoring tool in NAFLD for use as an outcome measure in clinical trial[60]. This tool known as the NAFLD Activity Score separately assigns severity of steatosis (0–3), inflammation (0–3) and ballooning (0–2) to give a cumulative score. Fibrosis is scored separately by the NAFLD Activity Score with 0 indicating no fibrosis, F1 some pericentral fibrosis (1b is periportal fibrosis), through F2 and F3 (bridging fibrosis) to F4 nodular change or cirrhotic change. Although children were used in the initial discovery cohort, it has since become clear that the pattern of disease in many children is distinct and that the NAFLD Activity Score does not reflect these features.

CYP frequently though are found to have type-2 disease which is a more periportal tract based pattern[61]. Fifty to seventy percent have a type-2 pattern or a cross-over between type 1 and type 2[62–64]. Type-2 disease has been studies in the context of severity, and both adults and children with histology more likely to have higher stage of fibrosis[65,66]. It is not clear if this pattern is due to a separate pathophysiological mechanism, although it certainly seems to be a marker of more advanced NASH.

The pathophysiology of why the location of inflammation and fibrosis varies is not understood. One possibility which may in part explain the preferential distribution is the concept of zonation. Along the liver lobe, the hepatocytes have different functions depending on their location in the lobule. For example, periportal hepatocytes are functionally specialised in Kreb's cycle amongst other tasks and those in the area of the central vein are rich in cytochrome P450 enzymes. The exposure of the liver to dietary components is more marked in zone 1 than in zone 3 (pericentral)[67].

It is likely that there is an evolutionary or a developmental reason for this localisation. It is possible that the repair mechanism in the liver, or rather the propensity and vigour with which the liver regenerates is different according to maturity and developmental stage. Fibroblast-specific protein 100, a fibroblast marker, has increased expression in interlobular ductal cells in paediatric control (healthy) liver than adult control liver, demonstrating that ductal epithelial cells in children have a greater tendency to exhibit features of mesenchymal cells and implying epithelial mesenchymal transition is more active within portal tracts in children[68,69].

An elegant body of work by Anna Mae Diehl and colleagues describe the localisation of hedgehog (Hh)

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signalling in the periportal region[69,70]. Hh signalling is expected to be more active in children. The Hh pathway is a pivotal morphogenic signalling pathway central in organogenesis. The pathway becomes quiescent in the liver during adolescence and reactivates in the presence of injury. Thus Hh signalling is involved in the activation of the regenerative pathway of liver. Healthy paediatric livers demonstrate more Hh signalling than healthy adult livers. Increased exposure to Hh ligands stimulated cells involved in wound healing. Normally these cells are tightly regulated however when deregulated, chronic inflammation, fibrosis and liver cancer may result. If children possess a proportionally greater number of Hh ligand producing cells and Hh responsive cells, then children may be at particular risk from insults that promote activation of the Hh pathway[70]. Guy *et al.* examined the association of portal fibrosis with the activation of the regenerative Hh signalling[71]. The number of Hh responsive cells correlated closely with both portal injury and the severity of fibrosis in liver biopsies of adult's patients with NAFLD ($P < 0{\cdot}0001$) supporting a relationship between Hh activation and liver damage. For this study, immunohistochemistry for Hh-ligand and Gli1 (Hh responsive cells) was undertaken on ninety biopsies within the NASH Clinical Research Network repository. A follow-on study in children again demonstrated more Hh activation with good correlation of Hh ligand expression and Hh responsive cells to liver injury and severity of disease[70].

In contrast, adult livers have more active pericentral Wnt signalling which is involved in glutamine synthesis, drug metabolism, bile acid and haem synthesis, regeneration and the response to oxidative stress[72].

Natural history and targeting intervention

The natural history of paediatric NAFLD has not yet been well described. Case series including one of 20 years describe occasional need for transplantation in young adulthood[72,73], but the rate of progression over years is not known[74]. Paired liver biopsies were analysed from 122 children who had enrolled in the placebo arm of two randomised clinical trials in NAFLD. Placebo groups were given lifestyle counselling for a period of 52 or 96 weeks. Over this time, fibrosis progressed in 23 % and improved in 34 %[75]. In children who present in the pre-teenage years, already established with stage 2–3 fibrosis, the rate of progression may be accelerated[76]. The heterogeneity within the population is not yet well understood but variability in phenotype may be due to underlying genetic susceptibility rather than environmental exposure.

The relative histological severity at presentation in children with this disease and the fact that alcohol is an unlikely confounding factor, means that paediatric NAFLD serves as an excellent disease model in evaluating pathophysiological mechanisms of development and thus targeting intervention in predisposed individuals.

Lifestyle change resulting in weight loss is an effective way of reversing or stabilising disease. In a meta-analysis of adults with NAFLD, weight loss of 5 % or more resulting in improvement in steatosis whereas ≥7 % weight loss resulted in improvement in steatohepatitis and in those with ≥10 % weight loss, all features of NAFLD were reversed or stabilised[77]. In a prospective study again in adults these outcomes were confirmed[78]. Only 50 % of the cohort were able to achieve 7 % of weight loss or more though of note in 94 % of those who achieved ≥5 % weight loss, fibrosis stabilised or reversed.

A small number of trials in children have demonstrated similar results. In an Italian study of eighty-four children, weight loss (average 4 kg) over a 12 months period achieved an improvement in alanine aminotransferase and steatosis on ultrasound[63]. Of the eighty-four children, fifty-seven (70 %) children completed the 12 month intervention with a mean 8 (SD 4·7) % decrease in weight in the fifty two who were overweight or obese. In the remaining five children who completed the study and had a BMI <85th centile, weight remained unchanged but alanine aminotransferase levels improved in two and normalised in three patients. Another paediatric study of intensive lifestyle intervention in North America achieved improvement in BMI *z*-score with a decrease of 0·1 U ($P < 0{\cdot}05$) baseline to 1 year and decrease in alanine aminotransferase in 69 % of the follow-up cohort. There was a 53 % drop-out rate however[79].

Insulin resistance is well recognised as an accompanying feature in 70 % of those with NAFLD, although not clearly associated with more or less severe disease in terms of inflammation and fibrosis. Insulin sensitisers have been studied frequently in clinical trial but without a consensus as to their effectiveness. In adults, the PIVENS trial was a randomised controlled trial of pioglitazone, vitamin E and placebo in treatment of adults with NASH but without type-2 diabetes over 96 weeks[80]. The specified outcome measure of improvement in the NAFLD Activity Score by ≥2 points was not reached with significance using pioglitazone. In children the TONIC trial compared metformin, vitamin E and placebo in 173 children with biopsy proven NAFLD[81]. Again, there was no statistically significant difference in the outcome measure (improvement in alanine aminotransferase) in those treated with metformin compared to those with placebo. Interestingly a change in homoeostatic model assessment-insulin resistance was not seen in those treated with metformin suggesting perhaps that treatment dose may have been insufficient or that selecting out those with insulin resistance may be more appropriate. The likelihood is that as the disease is relatively heterogeneous, treatments may need to be individually tailored.

Oxidative stress, most likely mediated by accumulation of fat droplets and the low grade inflammatory response accompanying visceral adiposity in the setting of genetic predisposition, is known to occur and perpetuate injury in NAFLD. Antioxidants, most commonly vitamin E have been trialled. Vitamin E has been shown in PIVENS to reduce steatohepatitis[82] and in TONIC to reduce ballooning. A significant difference in the main outcome measure (alanine aminotransferase) in the paediatric study TONIC was not found however[81].

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Current recommendation in adult practice is to use vitamin E in non-diabetic patients with biopsy proven NASH[83]. The PIVENS study has not yet been validated adequately however and thus caution is advised generally. There is no consensus on vitamin E use in children with NAFLD.

Ursodeoxycholic acid is another antioxidant which has been used in trial though without consistent effects. Cysteamine bitartrate works by increasing glutathione synthesis. This was used in the CyNCh trial in children with NAFLD in comparison to placebo over 52 weeks. Although there was an improvement in alanine aminotransferase, the primary outcome of histological improvement was not achieved with statistical significance[84].

The gut microbiome and its influence on bile acid metabolism is a new and emerging area in NAFLD and one which has led to the development of a cluster of new potential therapeutic agents. It is now recognised that the consumption of obesogenic foods leads to a change in the microbiota of the gut and to an increased permeability of the intestinal epithelium[51]. The microbiota influence the development of NAFLD through several different mechanisms among which is the production of SCFA which stimulate *de novo* triglyceride synthesis in the liver, modulation of choline metabolism (involved in VLDL synthesis), lipopolysaccharide production and of course modulation of bile acid metabolism[85].

The association between obesity and gut microbial change is well established in the form of a lower diversity of microbiota, although it is not yet clear whether this is a causal or a secondary effect. The change in microbial species leads to a higher concentration of secondary bile acids in the enterohepatic recirculation. In healthy individuals, bile acids play an important role in glucose and lipid metabolism regulating the negative feedback loops. Bile acids act via cellular receptors including farnesoid X receptor and G protein-coupled bile acid receptor to affect metabolism[85,86].

Probiotics and prebiotics are an alternative way to attempt to re-establish a healthy diversity of flora however there is no clear guidance on specific formulation or dose[85]. Weight loss, whether dietary induced or following bariatric surgery has been found to have a similar effect. Other important ways to disrupt the enterohepatic recirculation of bile thus decreasing the total bile pool is by using farnesoid X receptor agonists[87].

The process of fibrosis is in itself the common end point of several processes; steatosis with oxidative stress, inflammation and apoptosis. Prevention of fibrosis is optimal although antifibrotics are entering the clinical trial arena.

Although there is major interest in the search for the magic bullet for NAFLD, in reality the heterogeneity of the condition means that individualised treatment is needed, for example a patient with markers of increased oxidative stress may respond better to an antioxidant whereas those who have a poorly diverse microbiome may respond better to its modulation.

Everything considered, the most effective treatment (reversal) of NASH/steato-fibrosis is weight loss.

As discussed, there is evidence to show that interventions resulting in 5–10 % weight loss results in improvement in NASH[77]. In >50% however, this, weight loss is not achieved. The barriers to this weight loss need to be addressed. In children, weight loss *per se* may not be necessary as they grow into their centile. Thus, it is difficult to be prescriptive regarding the quantity of weight that needs to be lost and often CYP may require a BMI *z*-score to suit their specific genetic predisposition. Interestingly in a recent study of lifestyle intervention, there was a greater improvement in steatosis with weight loss in those with a higher genetic risk profile[88].

The importance of recognising mental health

Why does lifestyle intervention not work for all? Adherence may be the principal challenge and given that CYP are almost entirely dependent on family being involved in their goal, this increases complexity of the problem.

Social deprivation, a lack of education about the importance of a healthy lifestyle and cost of fresh, unprocessed foods are all important considerations and not all which can be successfully managed in the context of a clinical lifestyle intervention programme.

An underexplored area in ability to undertake and maintain lifestyle change in this population is the importance of mental health. The prevalence of depression and anxiety in CYP and adults with obesity is significant[89,90]. Depression is over-represented in adults with NAFLD as demonstrated in survey of 567 patients as part of the NASH Clinical Research Network with 53 % exhibiting subclinical depression and 14 % clinical depression, 45 % with subclinical anxiety with 25 % clinical anxiety. There were some histological correlates with presence of depression[91]. A small number of studies have shown that quality of life in both adults and children with NAFLD are significantly inferior to normal controls. In a survey of 239 children apart of the NASH Clinical Research Network, children with NAFLD had worse total physical and psychological quality of life scores as determined by the PedsQL questionnaire. Fatigue, trouble sleeping and sadness accounted for almost half the variance in quality of life scores[92]. In adults, degree of tiredness reported by patients with NAFLD was similar to that in women with primary biliary cirrhosis, a condition in which fatigue is often profound[93]. Although obstructive sleep apnoea may contribute to daytime somnolence in obesity, non-obstructive sleep apnoea sleep problems and the pathogenesis of this fatigue has not yet been systematically studied in NAFLD.

Response to lifestyle intervention in NAFLD is significantly less effective in the presence of depression particularly in the presence of acute major depressive disorder[94]. Although some studies in adults have used a psychological approach to lifestyle change in obesity related disorders, no trials in children are reported[95–97]. This approach included counselling sessions and cognitive behavioural therapy.

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Both adult and paediatric studies have drawn a link between depression, inflammation and the coexistence of complications of obesity, most frequently visceral adiposity associated conditions[98–100]. When combined with predisposing factors, immune challenges can lead to exaggerated or prolonged inflammatory responses[98]. The resulting sickness behaviours, depressive symptoms and poor lifestyle choices may lead to further inflammation. This pattern suggests that effective treatment options which target this vicious cycle may halt both the amplified inflammation and depressive symptoms.

Conclusions

The recognition of NAFLD in children is important in order to prevent progressive disease through young adulthood. Although closely associated with overweight and obesity, genetic and epigenetic clearly lead to susceptibility to the disease in the setting of an unhealthy lifestyle. At presentation 15 % of children will have bridging fibrosis and thus a relatively severe degree of liver injury. Liver injury is reversible however, and lifestyle change which addresses a mismatch in energy balance, is fundamental but achievable in less than 50 % of those undergoing an intensive programme with dietary advice and support. The reasons for this are as yet unclear but in part may be mediated by the high prevalence of depression and anxiety in those with this disease. There is a growing industry in treatments which target one of more pathophysiological process involved. Stratifying patients with NAFLD according to their susceptibilities and recognising comorbid mental health problems will facilitate individualised therapy and hopefully more successful outcomes.

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References

1. Cholankeril G, Wong RJ, Hu M *et al.* (2017) Liver transplantation for nonalcoholic steatohepatitis in the US: temporal trends and outcomes. *Dig Dis Sci* **62**, 2915–2922.
2. Williams R, Aspinall R, Bellis M *et al.* (2014) Addressing liver disease in the UK: a blueprint for attaining excellence in health care and reducing premature mortality from lifestyle issues of excess consumption of alcohol, obesity, and viral hepatitis. *Lancet* **384**, 1953–1997.
3. Estes C, Razavi H, Loomba R *et al.* (2018) Modeling the epidemic of nonalcoholic fatty liver disease demonstrates an exponential increase in burden of disease. *Hepatology* **67**, 123–133.
4. Ludwig J, Viggiano TR, McGill DB *et al.* (1980) Nonalcoholic steatohepatitis: Mayo Clinic experiences with a hitherto unnamed disease. *Mayo Clin Proc* **55**, 434–438.
5. Hardy T, Oakley F, Anstee QM *et al.* (2016) Nonalcoholic fatty liver disease: pathogenesis and disease spectrum. *Annu Rev Pathol* **11**, 451–496.
6. Hegarty R, Deheragoda M, Fitzpatrick E *et al.* (2018) Paediatric fatty liver disease (PeFLD): all is not NAFLD – pathophysiological insights and approach to management. *J Hepatol* **68**, 1286–1299.
7. Angulo P, Kleiner DE, Dam-Larsen S *et al.* (2015) Liver fibrosis, but no other histologic features, is associated with long-term outcomes of patients with nonalcoholic fatty liver disease. *Gastroenterology* **149**, 389–397, e10.
8. Ekstedt M, Franzen LE, Mathiesen UL *et al.* (2006) Long-term follow-up of patients with NAFLD and elevated liver enzymes. *Hepatology* **44**, 865–873.
9. Musso G, Gambino R, Cassader M *et al.* (2011) Meta-analysis: natural history of non-alcoholic fatty liver disease (NAFLD) and diagnostic accuracy of non-invasive tests for liver disease severity. *Ann Med* **43**, 617–649.
10. Younossi Z, Anstee QM, Marietti M *et al.* (2018) Global burden of NAFLD and NASH: trends, predictions, risk factors and prevention. *Nat Rev Gastroenterol Hepatol* **15**, 11–20.
11. Wattacheril J & Sanyal AJ (2016) Lean NAFLD: an underrecognized outlier. *Curr Hepatol Rep* **15**, 134–139.
12. Fitzpatrick E & Dhawan A (2016) Paediatric NAFLD: A distinct disease with the propensity for progressive fibrosis. In *Clinical Dilemmas in Non-Alcoholic Fatty Liver Disease*, pp. 29–35 [R Williams and SD Taylor-Robinson, editors]. Chichester: Wiley-Blackwell.
13. Quaglia A, Alves VA, Balabaud C *et al.* (2016) Role of aetiology in the progression, regression, and parenchymal remodelling of liver disease: implications for liver biopsy interpretation. *Histopathology* **68**, 953–967.
14. Anderson EL, Howe LD, Jones HE *et al.* (2015) The prevalence of non-alcoholic fatty liver disease in children and adolescents: a systematic review and meta-analysis. *PLoS One* **10**, e0140908.
15. Xanthakos SA, Jenkins TM, Kleiner DE *et al.* (2015) High prevalence of nonalcoholic fatty liver disease in adolescents undergoing bariatric surgery. *Gastroenterology* **149**, 623–634, e8.
16. Schwimmer JB, Deutsch R, Kahen T *et al.* (2006) Prevalence of fatty liver in children and adolescents. *Pediatrics* **118**, 1388–1393.
17. Xanthakos S, Miles L, Bucuvalas J *et al.* (2006) Histologic spectrum of nonalcoholic fatty liver disease in morbidly obese adolescents. *Clin Gastroenterol Hepatol* **4**, 226–232.
18. Lawlor DA, Callaway M, Macdonald-Wallis C *et al.* (2014) Nonalcoholic fatty liver disease, liver fibrosis, and cardiometabolic risk factors in adolescence: a cross-sectional study of 1874 general population adolescents. *J Clin Endocrinol Metab* **99**, E410–E417.

https://doi.org/10.1017/S0029665118002914 Published online by Cambridge University Press

19. Molleston JP, Schwimmer JB, Yates KP *et al.* (2014) Histological abnormalities in children with nonalcoholic fatty liver disease and normal or mildly elevated alanine aminotransferase levels. *J Pediatr* **164**, 707–713, e3.
20. Vos MB, Abrams SH, Barlow SE *et al.* (2017) NASPGHAN clinical practice guideline for the diagnosis and treatment of nonalcoholic fatty liver disease in children: recommendations from the expert committee on NAFLD (ECON) and the North American Society of Pediatric Gastroenterology, Hepatology and Nutrition (NASPGHAN). *J Pediatr Gastroenterol Nutr* **64**, 319–334.
21. Vajro P, Lenta S, Socha P *et al.* (2012) Diagnosis of nonalcoholic fatty liver disease in children and adolescents: position paper of the ESPGHAN Hepatology Committee. *J Pediatr Gastroenterol Nutr* **54**, 700–713.
22. Fitzpatrick E & Dhawan A (2014) Noninvasive biomarkers in non-alcoholic fatty liver disease: current status and a glimpse of the future. *World J Gastroenterol* **20**, 10851–10863.
23. Fitzpatrick E, Quaglia A, Vimalesvaran S *et al.* (2013) Transient elastography is a useful noninvasive tool for the evaluation of fibrosis in paediatric chronic liver disease. *J Pediatr Gastroenterol Nutr* **56**, 72–76.
24. Nobili V, Vizzutti F, Arena U *et al.* (2008) Accuracy and reproducibility of transient elastography for the diagnosis of fibrosis in pediatric nonalcoholic steatohepatitis. *Hepatology* **48**, 442–448.
25. Xanthakos SA, Podberesky DJ, Serai SD *et al.* (2014) Use of magnetic resonance elastography to assess hepatic fibrosis in children with chronic liver disease. *J Pediatr* **164**, 186–188.
26. Kaswala DH, Lai M & Afdhal NH (2016) Fibrosis Assessment in Nonalcoholic Fatty Liver Disease (NAFLD) in 2016. *Dig Dis Sci* **61**, 1356–1364.
27. Alkhouri N, Mansoor S, Giammaria P *et al.* (2014) The development of the pediatric NAFLD fibrosis score (PNFS) to predict the presence of advanced fibrosis in children with nonalcoholic fatty liver disease. *PLoS One* **9**, e104558.
28. Anstee QM, Seth D & Day CP (2016) Genetic factors that affect risk of alcoholic and nonalcoholic fatty liver disease. *Gastroenterology* **150**, 1728–1744, e7.
29. Goyal NP & Schwimmer JB (2018) The genetics of pediatric nonalcoholic fatty liver disease. *Clin Liver Dis* **22**, 59–71.
30. Speliotes EK, Yerges-Armstrong LM, Wu J *et al.* (2011) Genome-wide association analysis identifies variants associated with nonalcoholic fatty liver disease that have distinct effects on metabolic traits. *PLoS Genet* **7**, e1001324.
31. Romeo S, Kozlitina J, Xing C *et al.* (2008) Genetic variation in PNPLA3 confers susceptibility to nonalcoholic fatty liver disease. *Nat Genet* **40**, 1461–1465.
32. Chambers JC, Zhang W, Sehmi J *et al.* (2011) Genome-wide association study identifies loci influencing concentrations of liver enzymes in plasma. *Nat Genet* **43**, 1131–1138.
33. Romeo S, Sentinelli F, Cambuli VM *et al.* (2010) The 148M allele of the PNPLA3 gene is associated with indices of liver damage early in life. *J Hepatol* **53**, 335–338.
34. Santoro N, Zhang CK, Zhao H *et al.* (2012) Variant in the glucokinase regulatory protein (GCKR) gene is associated with fatty liver in obese children and adolescents. *Hepatology* **55**, 781–789.
35. Goffredo M, Caprio S, Feldstein AE *et al.* (2016) Role of TM6SF2 rs58542926 in the pathogenesis of nonalcoholic pediatric fatty liver disease: a multiethnic study. *Hepatology* **63**, 117–125.
36. Wattacheril J, Lavine JE, Chalasani NP *et al.* (2017) Genome-wide associations related to hepatic histology in nonalcoholic fatty liver disease in Hispanic boys. *J Pediatr* **190**, 100–107, e2.
37. Stender S, Kozlitina J, Nordestgaard BG *et al.* (2017) Adiposity amplifies the genetic risk of fatty liver disease conferred by multiple loci. *Nat Genet* **49**, 842–847.
38. Umano GR, Caprio S, Di Sessa A *et al.* (2018) The rs626283 variant in the MBOAT7 gene is associated with insulin resistance and fatty liver in Caucasian obese youth. *Am J Gastroenterol* **113**, 376–383.
39. Eslam M, Valenti L & Romeo S (2018) Genetics and epigenetics of NAFLD and NASH: clinical impact. *J Hepatol* **68**, 268–279.
40. Brumbaugh DE & Friedman JE (2014) Developmental origins of nonalcoholic fatty liver disease. *Pediatr Res* **75**, 140–147.
41. Wesolowski SR, Kasmi KC, Jonscher KR *et al.* (2017) Developmental origins of NAFLD: a womb with a clue. *Nat Rev Gastroenterol Hepatol* **14**, 81–96.
42. Maranghi F, Lorenzetti S, Tassinari R *et al.* (2010) In utero exposure to di-(2-ethylhexyl) phthalate affects liver morphology and metabolism in post-natal CD-1 mice. *Reprod Toxicol* **29**, 427–432.
43. Mouralidarane A, Soeda J, Visconti-Pugmire C *et al.* (2013) Maternal obesity programs offspring nonalcoholic fatty liver disease by innate immune dysfunction in mice. *Hepatology* **58**, 128–138.
44. Wankhade UD, Zhong Y, Kang P *et al.* (2017) Enhanced offspring predisposition to steatohepatitis with maternal high-fat diet is associated with epigenetic and microbiome alterations. *PLoS One* **12**, e0175675.
45. Lomas-Soria C, Reyes-Castro LA, Rodriguez-Gonzalez GL *et al.* (2018) Maternal obesity has sex-dependent effects on insulin, glucose and lipid metabolism and the liver transcriptome in young adult rat offspring. *J Physiol* **596**, 4611–4628.
46. McCurdy CE, Bishop JM, Williams SM *et al.* (2009) Maternal high-fat diet triggers lipotoxicity in the fetal livers of nonhuman primates. *J Clin Invest* **119**, 323–335.
47. Patel KR, White FV & Deutsch GH (2015) Hepatic steatosis is prevalent in stillborns delivered to women with diabetes mellitus. *J Pediatr Gastroenterol Nutr* **60**, 152–158.
48. Brumbaugh DE, Tearse P, Cree-Green M *et al.* (2013) Intrahepatic fat is increased in the neonatal offspring of obese women with gestational diabetes. *J Pediatr* **162**, 930–936, e1.
49. Poston L, Calevachetty R, Cnattingius S *et al.* (2016) Preconceptional and maternal obesity: epidemiology and health consequences. *Lancet Diabetes Endocrinol* **4**, 1025–1036.
50. Newton KP, Feldman HS, Chambers CD *et al.* (2017) Low and high birth weights are risk factors for nonalcoholic fatty liver disease in children. *J Pediatr* **187**, 141–146, e1.
51. Kalliomaki M, Collado MC, Salminen S *et al.* (2008) Early differences in fecal microbiota composition in children may predict overweight. *Am J Clin Nutr* **87**, 534–538.
52. Ayonrinde OT, Oddy WH, Adams LA *et al.* (2017) Infant nutrition and maternal obesity influence the risk of nonalcoholic fatty liver disease in adolescents. *J Hepatol* **67**, 568–576.
53. Vos MB, Kimmons JE, Gillespie C *et al.* (2008) Dietary fructose consumption among US children and adults: the Third National Health and Nutrition Examination Survey. *Medscape J Med* **10**, 160.
54. Sluik D, Engelen AI & Feskens EJ (2015) Fructose consumption in the Netherlands: the Dutch National Food Consumption Survey 2007–2010. *Eur J Clin Nutr* **69**, 475–481.

https://doi.org/10.1017/S0029665118002914 Published online by Cambridge University Press

55. Vos MB & Lavine JE (2013) Dietary fructose in non-alcoholic fatty liver disease. *Hepatology* **57**, 2525–2531.
56. Jin R, Willment A, Patel SS *et al.* (2014) Fructose induced endotoxemia in pediatric nonalcoholic Fatty liver disease. *Int J Hepatol* **2014**, 560620.
57. Mosca A, Nobili V, De Vito R *et al.* (2017) Serum uric acid concentrations and fructose consumption are independently associated with NASH in children and adolescents. *J Hepatol* **66**, 1031–1036.
58. Gibson PS, Lang S, Gilbert M *et al.* (2015) Assessment of diet and physical activity in paediatric non-alcoholic fatty liver disease patients: A United Kingdom Case Control Study. *Nutrients* **7**, 9721–9733.
59. Jin R, Le NA, Liu S *et al.* (2012) Children with NAFLD are more sensitive to the adverse metabolic effects of fructose beverages than children without NAFLD. *J Clin Endocrinol Metab* **97**, E1088–E1098.
60. Kleiner DE, Brunt EM, Van Natta M *et al.* (2005) Design and validation of a histological scoring system for non-alcoholic fatty liver disease. *Hepatology* **41**, 1313–1321.
61. Yeh MM & Brunt EM (2007) Pathology of nonalcoholic fatty liver disease. *Am J Clin Pathol* **128**, 837–847.
62. Fitzpatrick E, Mitry RR, Quaglia A *et al.* (2010) Serum levels of CK18 M30 and leptin are useful predictors of steatohepatitis and fibrosis in paediatric NAFLD. *J Pediatr Gastroenterol Nutr* **51**, 500–506.
63. Nobili V, Marcellini M, Devito R *et al.* (2006) NAFLD in children: a prospective clinical-pathological study and effect of lifestyle advice. *Hepatology* **44**, 458–465.
64. Schwimmer JB, Behling C, Newbury R *et al.* (2005) Histopathology of pediatric nonalcoholic fatty liver disease. *Hepatology* **42**, 641–649.
65. Brunt EM, Kleiner DE, Wilson LA *et al.* (2009) Portal chronic inflammation in nonalcoholic fatty liver disease (NAFLD): a histologic marker of advanced NAFLD-Clinicopathologic correlations from the nonalcoholic steatohepatitis clinical research network. *Hepatology* **49**, 809–820.
66. Africa JA, Behling CA, Brunt EM *et al.* (2018) In children with nonalcoholic fatty liver disease, zone 1 steatosis is associated with advanced fibrosis. *Clin Gastroenterol Hepatol* **16**, 438–446, e1.
67. Nobili V, Mosca A, De Vito R *et al.* (2018) Liver zonation in children with non-alcoholic fatty liver disease: Associations with dietary fructose and uric acid concentrations. *Liver Int* **38**, 1102–1109.
68. Omenetti A, Bass LM, Anders RA *et al.* (2011) Hedgehog activity, epithelial-mesenchymal transitions, and biliary dysmorphogenesis in biliary atresia. *Hepatology* **53**, 1246–1258.
69. Omenetti A, Choi S, Michelotti G *et al.* (2011) Hedgehog signaling in the liver. *J Hepatol* **54**, 366–373.
70. Swiderska-Syn M, Suzuki A, Guy CD *et al.* (2013) Hedgehog pathway and pediatric nonalcoholic fatty liver disease. *Hepatology* **57**, 1814–1825.
71. Guy CD, Suzuki A, Abdelmalek MF *et al.* (2015) Treatment response in the PIVENS trial is associated with decreased Hedgehog pathway activity. *Hepatology* **61**, 98–107.
72. Gebhardt R & Matz-Soja M (2014) Liver zonation: novel aspects of its regulation and its impact on homeostasis. *World J Gastroenterol* **20**, 8491–8504.
73. Molleston JP, White F, Teckman J *et al.* (2002) Obese children with steatohepatitis can develop cirrhosis in childhood. *Am J Gastroenterol* **97**, 2460–2462.
74. Feldstein AE, Charatcharoenwitthaya P, Treeprasertsuk S *et al.* (2009) The natural history of non-alcoholic fatty liver disease in children: a follow-up study for up to 20 years. *Gut* **58**, 1538–1544.
75. Xanthakos S, Lavine J, Yates K *et al.* (2017) Natural history of non alcoholic fatty liver disease in children receiving standard lifestyle counselling and placebo in NASH Clinical Research Network trials. *Hepatology* **66** 1 supp, 31A.
76. Goh GB & McCullough AJ (2016) Natural history of non-alcoholic fatty liver disease. *Dig Dis Sci* **61**, 1226–1233.
77. Musso G, Cassader M, Rosina F *et al.* (2012) Impact of current treatments on liver disease, glucose metabolism and cardiovascular risk in non-alcoholic fatty liver disease (NAFLD): a systematic review and meta-analysis of randomised trials. *Diabetologia* **55**, 885–904.
78. Vilar-Gomez E, Martinez-Perez Y, Calzadilla-Bertot L *et al.* (2015) Weight loss through lifestyle modification significantly reduces features of nonalcoholic steatohepatitis. *Gastroenterology* **149**, 367–378, e5; quiz e14-5.
79. DeVore S, Kohli R, Lake K *et al.* (2013) A multidisciplinary clinical program is effective in stabilizing BMI and reducing transaminase levels in pediatric patients with NAFLD. *J Pediatr Gastroenterol Nutr* **57**, 119–123.
80. Chalasani NP, Sanyal AJ, Kowdley KV *et al.* (2009) Pioglitazone versus vitamin E versus placebo for the treatment of non-diabetic patients with non-alcoholic steatohepatitis: PIVENS trial design. *Contemp Clin Trials* **30**, 88–96.
81. Lavine JE, Schwimmer JB, Van Natta ML *et al.* (2011) Effect of vitamin E or metformin for treatment of non-alcoholic fatty liver disease in children and adolescents: the TONIC randomized controlled trial. *JAMA* **305**, 1659–1668.
82. Sanyal AJ, Chalasani N, Kowdley KV *et al.* (2010) Pioglitazone, vitamin E, or placebo for nonalcoholic steatohepatitis. *N Engl J Med* **362**, 1675–1685.
83. Chalasani N, Younossi Z, Lavine JE *et al.* (2018) The diagnosis and management of nonalcoholic fatty liver disease: practice guidance from the American Association for the Study of Liver Diseases. *Hepatology* **67**, 328–357.
84. Schwimmer JB, Lavine JE, Wilson LA *et al.* (2016) In children with nonalcoholic fatty liver disease, cysteamine bitartrate delayed release improves liver enzymes but does not reduce disease activity scores. *Gastroenterology* **151**, 1141–1154, e9.
85. Chen J, Thomsen M & Vitetta L (2019) Interaction of gut microbiota with dysregulation of bile acids in the pathogenesis of nonalcoholic fatty liver disease and potential therapeutic implications of probiotics. *J Cell Biochem* **120**, 2713–2720.
86. Kuipers F, Bloks VW & Groen AK (2014) Beyond intestinal soap – bile acids in metabolic control. *Nat Rev Endocrinol* **10**, 488–498.
87. Neuschwander-Tetri BA, Loomba R, Sanyal AJ *et al.* (2015) Farnesoid X nuclear receptor ligand obeticholic acid for non-cirrhotic, non-alcoholic steatohepatitis (FLINT): a multicentre, randomised, placebo-controlled trial. *Lancet* **385**, 956–965.
88. Ma J, Hennein R, Liu C *et al.* (2018) Improved diet quality associates with reduction in liver fat, particularly in individuals with high genetic risk scores for nonalcoholic fatty liver disease. *Gastroenterology* **155**, 107–117.
89. Leon G, de Klerk E, Ho J *et al.* (2018) Prevalence of comorbid conditions pre-existing and diagnosed at a tertiary care pediatric weight management clinic. *J Pediatr Endocrinol Metab* **31**, 385–390.
90. Mannan M, Mamun A, Doi S *et al.* (2016) Prospective associations between depression and obesity for adolescent

https://doi.org/10.1017/S0029665118002914 Published online by Cambridge University Press

males and females- a systematic review and meta-analysis of longitudinal studies. *PLoS One* **11**, e0157240.
91. Youssef NA, Abdelmalek MF, Binks M *et al.* (2013) Associations of depression, anxiety and antidepressants with histological severity of nonalcoholic fatty liver disease. *Liver Int* **33**, 1062–1070.
92. Kistler KD, Molleston J, Unalp A *et al.* (2010) Symptoms and quality of life in obese children and adolescents with non-alcoholic fatty liver disease. *Aliment Pharmacol Ther* **31**, 396–406.
93. Newton JL, Jones DE, Henderson E *et al.* (2008) Fatigue in non-alcoholic fatty liver disease (NAFLD) is significant and associates with inactivity and excessive daytime sleepiness but not with liver disease severity or insulin resistance. *Gut* **57**, 807–813.
94. Tomeno W, Kawashima K, Yoneda M *et al.* (2015) Non-alcoholic fatty liver disease comorbid with major depressive disorder: the pathological features and poor therapeutic efficacy. *J Gastroenterol Hepatol* **30**, 1009–1014.
95. Gelli C, Tarocchi M, Abenavoli L *et al.* (2017) Effect of a counseling-supported treatment with the Mediterranean diet and physical activity on the severity of the non-alcoholic fatty liver disease. *World J Gastroenterol* **23**, 3150–3162.
96. Moscatiello S, Di Luzio R, Bugianesi E *et al.* (2011) Cognitive-behavioral treatment of nonalcoholic Fatty liver disease: a propensity score-adjusted observational study. *Obesity (Silver Spring)* **19**, 763–770.
97. Bellentani S, Dalle Grave R, Suppini A *et al.* (2008) Behavior therapy for nonalcoholic fatty liver disease: the need for a multidisciplinary approach. *Hepatology* **47**, 746–754.
98. Kiecolt-Glaser JK, Derry HM & Fagundes CP (2015) Inflammation: depression fans the flames and feasts on the heat. *Am J Psychiatry* **172**, 1075–1091.
99. Silva N, Atlantis E & Ismail K (2012) A review of the association between depression and insulin resistance: pitfalls of secondary analyses or a promising new approach to prevention of type 2 diabetes? *Curr Psychiatry Rep* **14**, 8–14.
100. Byrne ML, O'Brien-Simpson NM, Mitchell SA *et al.* (2015) Adolescent-Onset Depression: Are Obesity and Inflammation Developmental Mechanisms or Outcomes? *Child Psychiatry Hum Dev* **46**, 839–850.

https://doi.org/10.1017/S0029665118002914 Published online by Cambridge University Press