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Risk and protective factors of non-alcoholic fatty liver disease in paediatric obesity: A nationwide nested case–control study

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Summary

Not all children with obesity carry a similar risk of non-alcoholic fatty liver disease (NAFLD). We investigated the effect of obesity severity, metabolic risk parameters, and obesity treatment outcome on later risk of NAFLD in paediatric obesity. We conducted a nested case–control study of children and adolescents enrolled in the Swedish Childhood Obesity Treatment Register (BORIS) (2001–2016). NAFLD was ascertained from the National Patient Register. Five controls per case were matched by sex and age at index date and at the obesity treatment initiation. Seventy-six pairs (n cases = 76, n controls = 241) were included in the analysis (29% females, mean age at obesity treatment initiation was 10.8 ± 3.07 years). Mean age of NAFLD diagnosis was 14.2 ± 3.07 years. The risk for NAFLD increased with severe obesity (odds ratio [OR]: 3.15, 95% confidence interval [CI]: 1.69–5.89), impaired fasting glucose (OR: 5.29, 95% CI: 1.40–20.06), high triglycerides (OR: 2.33, 95% CI: 1.22–4.43) and weight gain (OR: 4.67, 95% CI: 1.51–14.49 per body mass index standard deviation score [BMI SDS] unit). Relative weight loss of at least 0.25 BMI SDS units reduced NAFLD risk independently of other risk factors (OR: 0.09, 95% CI: 0.01–0.56). Severe obesity, impaired fasting glucose and high triglycerides are risk factors for future NAFLD in paediatric obesity. Successful obesity treatment almost eliminates the risk for NAFLD independently of obesity severity, IFG and high triglycerides.

KEYWORDS
childhood obesity, epidemiology, fatty liver, metabolic diseases

What is already known about this subject

- Childhood obesity is associated with paediatric non-alcoholic fatty liver disease (NAFLD).
- Which children and adolescents with obesity having the greatest risk of NAFLD are not well understood.
- The effect of weight change on future risk of NAFLD among children and adolescents undergoing long-term obesity treatment is not known.

What this study adds

- Severe obesity, impaired fasting glucose and high triglycerides are risk factors for future NAFLD in paediatric obesity.

- Successful obesity treatment achieved in a long-term obesity treatment reduces the risk for NAFLD, while poor treatment outcome increases the risk.

1 | INTRODUCTION

Paediatric non-alcoholic fatty liver disease (NAFLD) encompasses steatosis, marked by infiltration of fat in the liver, to the progressive forms, such as steatohepatitis, fibrosis and cirrhosis.[1] Paediatric NAFLD is caused by a large spectrum of diseases, including genetic, metabolic and syndromic disorders.[2] Yet, paediatric NAFLD is often considered as an obesity-related comorbidity.[3] NAFLD has nowadays become one of the most common causes of chronic liver disease in children and adolescents,[2] with a reported prevalence of 28%–41% among those with obesity.[4]

To present, the pathogenesis of paediatric NAFLD has not been fully understood. Some longitudinal population-based studies have demonstrated obesity[5,6] and insulin resistance[7] as predisposing factors of NAFLD in children and adolescents. However, not all children and adolescents with obesity carry a similar risk of NAFLD.[8] Among paediatric obesity population, NAFLD is more prevalent among those with severe obesity[9–11] and those having impaired glucose metabolism.[9–13] Nevertheless, the direction of causality is still unclear due to the cross-sectional design used in the previous studies.[9–13]

Several interventions in children and adolescents with obesity have shown that weight loss improves serum alanine aminotransferase (ALT)[14,15] and decreases liver fat echogenicity.[16,17] Those interventions were conducted in weight loss camps[14,17] or using short-term structured weekly and monthly programmes.[15,16] Yet, the effect of weight loss on NAFLD risk among children and adolescents with obesity receiving long-term obesity treatment in clinical practice has not yet been established.

In the present study, we aimed to investigate the associations of obesity severity, metabolic risk parameters and obesity treatment outcome with later risk of NAFLD in a large cohort of children and adolescents with obesity.

2 | MATERIALS AND METHODS

2.1 | Study design and population

We performed a matched nested case–control study within a prospective cohort consisting of children and adolescents with obesity who started their obesity treatment between 2001 and 2016 and were enrolled in the Swedish Childhood Obesity Treatment Register (BORIS) ($n = 16\,291$). BORIS was initiated in 2005 and provides prospective documentation for long-term monitoring of childhood obesity treatment in Sweden. Retrospective data on children and adolescents who started obesity treatment between 1994 and 2005 have been entered. The treatment consists mainly of behavioural lifestyle. Visit frequency varies between individuals, but annual visits are recommended regardless of successful treatment. Patients and their guardians are informed about data collection in BORIS and are free to opt out from the register. Further details about BORIS are published elsewhere.[18]

This population was followed for NAFLD diagnosis until 31 December 2017. The inclusion criterion was all children and adolescents aged <18 years, who were diagnosed with obesity according to the International Obesity Task Force[19] at obesity treatment initiation. Index date was defined as the date of the first recorded NAFLD diagnosis.

To avoid mixed aetiologies of fatty liver disease, we excluded individuals diagnosed with: (a) viral or autoimmune hepatitis; (b) primary biliary cholangitis; (c) alcoholic liver disease; (d) type 1 diabetes mellitus; (e) liver glycogen storage disease; (f) Wilson's disease; (g) iron metabolism disorder; (h) glucose-6-phosphate dehydrogenase (G6PD) deficiency; (i) toxic liver disease; (j) hepatic failure; (k) the genetic syndromes, i.e., Down syndrome, Prader-Willi syndrome, Laurence-Moon-Bardet-Biedl syndrome, Russel-Silver syndrome, Klinefelter syndrome, Fragile-X syndrome, Turner syndrome; or (l) individuals prescribed systemic corticosteroid drug within 6 years prior to index date. Those without anthropometric and metabolic parameters data within 1–6 years prior to index date were also excluded (Figure 1). This study was approved by the Regional Ethical Review Board in Stockholm (2016/944-31).

2.2 | Data sources

In Sweden, every resident is assigned a unique personal identity number. Through the personal identity number, BORIS register was linked to the National Patient Register, the National Prescribed Medication Register and the Cause of Death Register. The National Patient Register contains data on clinical diagnoses of inpatient care (since 1987) and outpatient specialist care (since 2001). All clinical diagnoses are coded according to the International Classification of Diseases (ICD) system. The positive predictive value of ICD-10 codes in the National Patient Register is 85%–95% for most chronic diseases, with medical records and Swedish national quality registers as reference.[20]

2.3 | Selection of cases and controls

Cases were defined as any individuals who were diagnosed with NAFLD during the study period. NAFLD diagnosis was identified according to ICD-10 codes (K75.8, K75.81, K75.9, K76.0) recorded in the National Patient Register. All NAFLD diagnoses were established in specialist care settings. Most cases were diagnosed in paediatric centres (Table S1). In daily practice, the diagnosis of NAFLD in

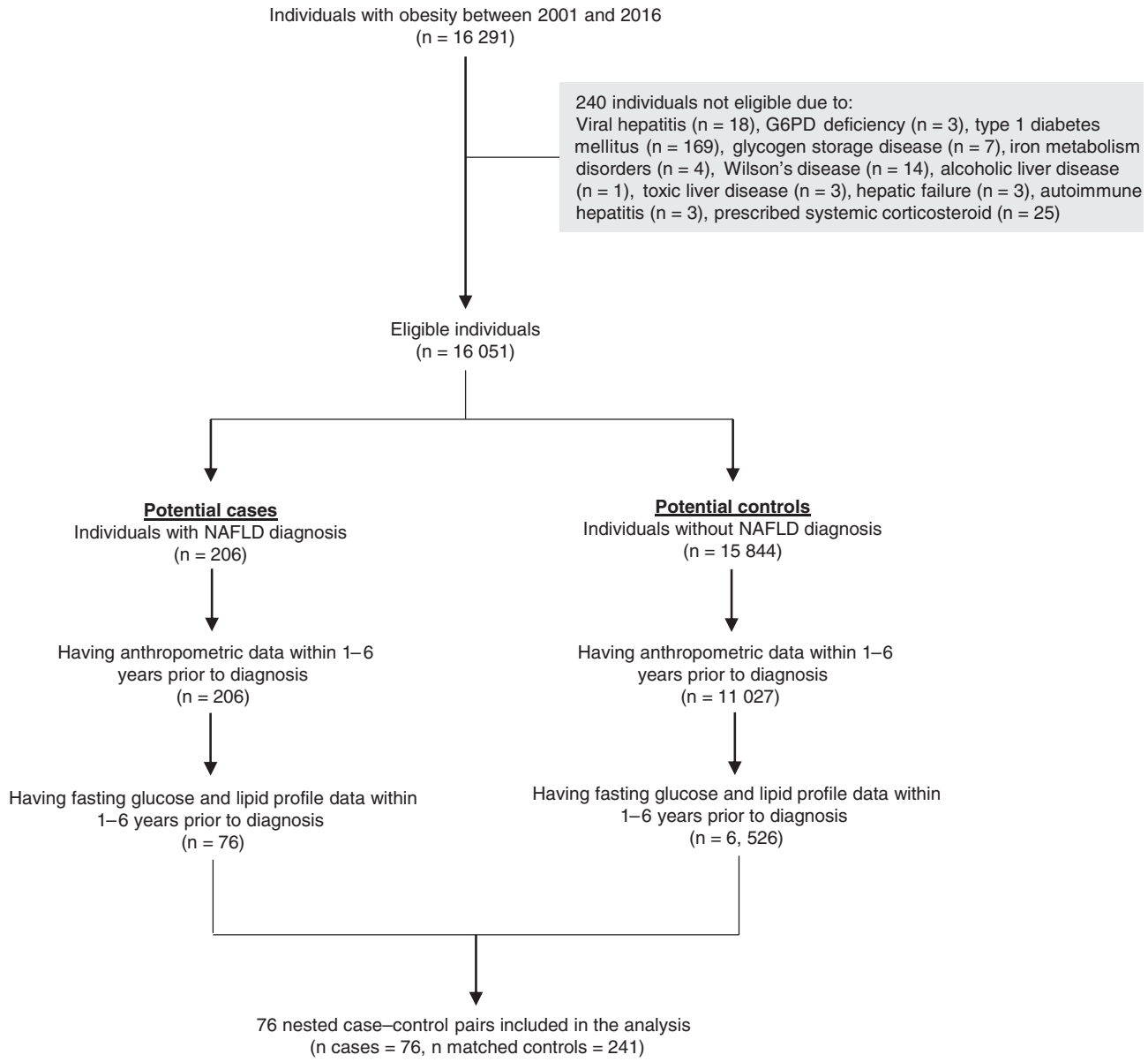

FIGURE 1 Flowchart of the exclusion process

children with obesity is often made by ultrasound which shows if fatty liver is present and other suspected causes of fatty liver have been ruled out. Cases were restricted as the first occurrence of NAFLD diagnosis. Of 206 individuals having NAFLD diagnosis, 76 cases were included in the analysis (Figure 1).

Controls without recorded NAFLD diagnosis were randomly selected from the study population. For each case, up to five eligible controls were matched by the following criteria: (i) age at index date (±1 year); (ii) sex; (iii) age at the first visit of obesity treatment (±1 year); and (iv) age at the anthropometric measurement (±1 year). Case–control matching was performed without replacement. In addition, eligible controls were those who were alive at least as long as the cases. After the exclusion and matching process, 241 matched controls of 6526 available controls were included in the analysis (Figure 1).

2.4 | Assessment of exposures

To assess whether metabolic parameters during obesity treatment are associated with risk for later NAFLD in a causal direction, data on exposure variables were measured between 1 year and 6 years prior to the index date. The degree of obesity was measured using body mass index standard deviation score (BMI SDS).[19] BMI SDS was classified into three categories: overweight, obesity and severe obesity; equivalent to adult BMI of 25 to <30, 30 to <35, and ≥35 kg/m^2, respectively.[19] Impaired fasting glucose (IFG) was defined by fasting glucose between 6.1 and 6.9 mmol/L. As some cross-sectional studies have shown an association between paediatric NAFLD and lipid components,[21,22] we assessed the longitudinal association of lipid components and NAFLD in the present study. Lipid profile was classified as follows: "high total cholesterol" (total cholesterol ≥5.2 mmol/

L), "high LDL" (LDL ≥3.4 mmol/L), "low HDL" (HDL <1.0 mmol/L), "high triglycerides" (triglycerides ≥1.2 mmol/L in individuals aged 5–9 years or ≥1.6 mmol/L in individuals aged 10–18 years).[23] In addition, other metabolic parameter data (such as waist circumference, blood pressure, transaminases, fasting insulin and homeostasis model assessment [HOMA]) were available and assessed in a subset of individuals.

To assess the association of NAFLD with Nordic background, we classified the individuals as "Nordic" (individuals with at least one of their parents born in a Nordic country, i.e., Sweden, Finland, Denmark, Norway or Iceland) and "non-Nordic" (individuals born outside Nordic countries or born in a Nordic country with both parents born outside Nordic countries).

Obesity treatment outcome was measured by subtracting BMI SDS at the first visit from BMI SDS at the last visit prior to the index date. As a BMI SDS reduction of 0.25 or greater has previously been shown to improve cardiovascular risk factors in children with obesity,[24] we used this cut-off value to evaluate whether a BMI SDS reduction of 0.25 or greater also reduces the risk for NAFLD.

2.5 | Statistical analysis

Categorical data are presented as proportions. Continuous data are presented as means and standard deviations (SD) or median (percentile 25th [P25], percentile 75th [P75]), depending on the data distribution. Histogram was used to identify the shape of data distribution. The distribution of baseline and clinical characteristics of cases and controls was compared by using independent *t* test or Mann–Whitney *U* test for continuous variables, and chi-square test for categorical variables. In the analyses, degree of obesity categories was collapsed into two categories, i.e., overweight/obesity and severe obesity, since there were very few individuals ($n = 28$) with overweight within 1–6 years prior to index date.

TABLE 1 Baseline and clinical characteristics

	Cases	Matched controls	*p*-Value
Matching criteria			
Female, *n* (%)	21 (27.6)	70 (29.1)	.812
Age at first visit of obesity treatment (years), mean (SD)	10.8 (3.28)	10.8 (3.01)	.995
Age at metabolic parameters measurements (years), mean (SD)	11.5 (3.12)	11.5 (2.90)	.97
Age at index date (years), mean (SD)	14.3 (3.07)	14.7 (2.98)	.305
Nordic background			
Nordic, *n* (%)	50 (65.8)	157 (65.2)	.918
Non-Nordic, *n* (%)	26 (34.2)	84 (34.8)	
Metabolic parameters within 1–6 years prior to index date[a]			
BMI SDS, mean (SD)	3.11 (0.50)	2.78 (0.51)	<.001
Overweight, *n* (%)	2 (2.6)	26 (10.8)	<.001
Obesity, *n* (%)	24 (31.6)	122 (50.6)	
Severe obesity, *n* (%)	50 (65.8)	93 (38.6)	
Fasting glucose (mmol/L), mean (SD)[b]	5.25 (0.49)	5.16 (0.45)	.13
IFG, *n* (%)	5 (6.58)	6 (2.49)	.089
Total cholesterol (mmol/L), mean (SD)	4.48 (0.83)	4.27 (0.72)	.033
High total cholesterol, *n* (%)	11 (14.47)	30 (12.45)	.646
LDL (mmol/L), mean (SD)	2.80 (0.78)	2.60 (0.67)	.028
High LDL, *n* (%)	14 (18.42)	33 (13.69)	.312
HDL (mmol/L), mean (SD)	1.11 (0.26)	1.26 (0.35)	.0005
Low HDL, *n* (%)	23 (30.26)	42 (17.43)	.016
Triglycerides (mmol/L), mean (SD)	1.51 (0.87)	1.16 (0.65)	.0002
High triglycerides, *n* (%)	26 (34.21)	40 (16.60)	.001

Notes: *n* cases = 76, *n* controls = 241. *p*-Values were calculated using independent *t* test or chi-square test, where appropriate.
Abbreviations: BMI SDS, body mass index standard deviation score; HDL, high-density lipoprotein; IFG, impaired fasting glucose; LDL, low-density lipoprotein.
[a]Index date: date of non-alcoholic fatty liver disease diagnosis.
[b]There were no cases with fasting glucose ≥7.0 mmol/L, i.e., type 2 diabetes.

Crude and adjusted odds ratios for NAFLD were calculated using conditional logistic regression. As a result of matched case–control and conditional analyses, the crude analyses were controlled for sex, age at obesity treatment initiation, age at anthropometric measurement and age at index date. Since collinearity was detected between components of lipid profile, separate multivariate models for each component of lipid profile were performed. To assess the effect of obesity treatment on NAFLD, BMI SDS changes (BMI SDS at the last visit – BMI SDS at the first visit) were analysed as continuous and categorical variables. Statistical significance was defined as $p < .05$. Data were managed and analysed using STATA version 16 (StataCorp, College Station, TX, USA).

3 | RESULTS

3.1 | Baseline and clinical characteristics

Seventy-six nested case–control pairs (n cases = 76, n controls = 241) were included in the study. The flowchart is shown in Figure 1. Individuals with NAFLD included in the analysis were 2.5 years younger at their first visit of obesity treatment than individuals with NAFLD excluded from the analysis ($p < .001$). Neither sex, age at NAFLD diagnosis, BMI SDS nor degree of obesity differed between individuals with NAFLD included and excluded from the analysis (Table S2).

A total of 27.6% cases and 29.1% controls were females. The mean age at the obesity treatment initiation among all individuals was 10.7 (SD 3.1) years. The mean age at NAFLD diagnosis was 14.3 (SD 3.07) among cases. The median of ALT measured within 6 months prior to NAFLD diagnosis among cases was 84 U/L (P25 = 55 U/L, P 75 = 148 U/L, data available in $n = 37$) and among controls was 28 U/L (P25 = 19 U/L, P75 = 36 U/L, $n = 67$) ($p < .001$). As a result of matching, there were no significant differences of sex, age at the obesity treatment initiation, age at clinical parameters measurements and age at index date between cases and controls, $p > .05$ (Table 1).

Of the 76 individuals with NAFLD diagnosis, 74 (97.4%) were diagnosed with *fatty liver, not elsewhere classified* (ICD-10 code K76.0), and the others were diagnosed with *other specified inflammatory liver diseases* (K75.8).

The proportion of individuals of Nordic origin did not differ between cases and controls (65.8% vs. 65.2%, $p = .918$). Among the cases, 65.8% had severe obesity vs. 38.6% in the control group, $p < .001$. Clinical characteristics of cases and controls are presented in Table 1. In a subset of individuals, cases were found to have a higher proportion of hypertension and elevated transaminases, as well as a higher median fasting insulin and HOMA, compared to the controls (all p-values <.05) (Table S3).

3.2 | Association of NAFLD with metabolic parameters

In adjusted analyses, severe obesity was associated with a 3.2-fold increased risk of NAFLD (OR: 3.16, 95% CI 1.69 to 5.89, $p < .001$), and high level of triglycerides was associated with a 2.3-fold increased risk of NAFLD (OR: 2.33, 95% CI 1.22 to 4.43, $p = .010$). IFG was not associated with NAFLD in the crude analysis ($p = .056$); however, after adjustment for degree of obesity and presence of high triglycerides, IFG became a significant risk factor of NAFLD (OR: 5.30, 95% CI: 1.40 to 20.06, $p = .014$). On the contrary, the OR for low HDL was attenuated when it was adjusted for degree of obesity and IFG (OR for low HDL: 1.70, 95% CI: 0.89 to 3.24, $p = .110$). The adjusted odds ratios of the metabolic parameters are shown in Table 2.

3.3 | The impact of obesity treatment outcome on NAFLD

To assess the impact of obesity treatment outcome on NAFLD, individuals with early dropout, namely <1 year of obesity treatment, were excluded. A total of 54 matched pairs (54 cases and 118 controls) were included in the analyses. Individuals included in these analyses were 1.5 years younger at their first visit of obesity treatment than individuals with early dropout. Neither sex, age at NAFLD diagnosis or degree of obesity differed between individuals with ≥1 year of treatment and those with early dropout.

An increase in BMI SDS over time was associated with NAFLD (OR: 4.67 per BMI SDS unit, 95% CI: 1.51 to 14.49, $p = .007$), independently of degree of obesity, the presence of IFG and high triglycerides 1–6 years prior to index date. Compared to individuals without any BMI SDS changes, individuals with reduction in BMI SDS had lower risk of NAFLD (Figure 2).

When BMI SDS changes were analysed as categorical variable, 24.1% achieved a BMI SDS reduction of at least 0.25 unit among cases versus 44.1% among controls, $p = .014$. A BMI SDS reduction of at least 0.25 unit was associated with lower odds of future NAFLD (OR: 0.09, 95% CI: 0.01 to 0.56, $p = .010$) after adjustment for treatment duration, degree of obesity, presence of IFG and high

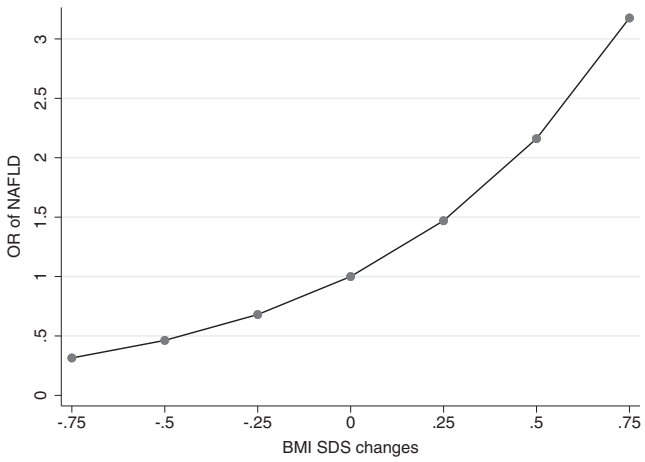

FIGURE 2 Odds ratios of non-alcoholic fatty liver disease (NAFLD) for body mass index standard deviation score (BMI SDS) changes adjusted for degree of obesity, the presence of impaired fasting glucose (IFG) and high triglycerides. OR, odds ratio

TABLE 2 Crude and adjusted odds ratios (ORs) of non-alcoholic fatty liver disease for metabolic parameters

	Crude		Adjusted	
Metabolic parameters	**OR (95% CI)**	***p*-Value**	**OR (95% CI)**	***p*-Value**
Severe obesity (vs. overweight and obesity)	3.40 (1.86–6.22)	<.001	3.15 (1.69–5.89)[a]	<.001
IFG (yes vs. no)	3.43 (0.97–12.11)	.056	5.29 (1.40–20.05)[b]	.014
High triglycerides (yes vs. no)	2.50 (1.38–4.51)	.01	2.33 (1.22–4.43)[c]	.010
Low HDL (yes vs. no)	2.18 (1.19–4.00)	.012	1.70 (0.89–3.24)[c]	.110
High LDL (yes vs. no)	1.20 (0.59–2.46)	.611	1.04 (0.50–2.20)[c]	.912
High total cholesterol (yes vs. no)	0.97 (0.45–2.08)	.938	0.85 (0.38–1.88)[c]	.683

Notes: *n* cases = 76, *n* controls = 241. Total cholesterol, HDL cholesterol, LDL cholesterol and triglyceride were correlated with each other. The adjusted analyses were therefore separated for each lipid profile. ORs and *p*-values were calculated using conditional logistic regression.
Abbreviations: CI, confidence interval; HDL, high-density lipoprotein; IFG, impaired fasting glucose; LDL, low-density lipoprotein; OR, odds ratio.
[a]Adjusted for presence of impaired fasting glucose and high triglycerides.
[b]Adjusted for degree of obesity and presence of high triglycerides.
[c]Adjusted for degree of obesity and presence of impaired fasting glucose.

TABLE 3 Associations of non-alcoholic fatty liver disease with obesity treatment outcome[a]

			Crude		Adjusted[b]	
	Cases	**Controls**	**OR (95% CI)**	***p*-Value**	**OR (95% CI)**	***p*-Value**
Reduction of at least 0.25 BMI SDS units, *n* (%)	13 (24.1)	52 (44.1)	0.33 (0.13–0.80)	.014	0.08 (0.01–0.56)	<.001
Treatment duration (months), median (min–max)	44 (14–137.5)	27 (12–110)	1.16 (1.09–1.22)	<.001	1.17 (1.09–1.27)	<.001

Abbreviation: BMI SDS, body mass index standard deviation score.
[a]Fifty-four matched pairs were included (*n* cases = 54, *n* controls = 118).
[b]Adjusted for degree of obesity, impaired fasting glucose, high triglycerides, reduction of at least 0.25 BMI SDS units and treatment duration.

triglycerides. The median of treatment duration was longer among the cases compared to the controls (44 months vs. 27 months, $p < .001$). In the adjusted analysis, OR of NAFLD for treatment duration was 1.17 (95% CI: 1.09 to 1.27, $p < .001$) for each month increase. The association between obesity treatment outcome, treatment duration and NAFLD are presented in Table 3.

Among the 54 matched pairs, the associations of IFG and high triglycerides with NAFLD were not significant when adjusted for reduction of at least 0.25 BMI SDS units, treatment duration and degree of obesity (OR for IFG: 14.22, 95% CI: 0.21 to 967.32, $p = .218$) (OR for high triglycerides: 1.66, 95% CI: 0.42 to 6.52, $p = .338$).

4 | DISCUSSION

This case–control study, which was nested in a large cohort of children and adolescents with obesity in Sweden, confirms theories from the previous cross-sectional studies[10–13,25,26] and provides evidence of longitudinal associations between obesity severity, the presence of IFG, high level of triglycerides and later risk of NAFLD. Moreover, the present study shows that individuals with relative weight loss of at least 0.25 BMI SDS units had a lower risk of future NAFLD, while BMI SDS gain was associated with an increased risk of NAFLD. No association between NAFLD and Nordic background was found.

The present study demonstrated that individuals with severe obesity had a triple risk of NAFLD compared to those with overweight or class 1 obesity. This association was independent of age, sex, IFG and high triglycerides. This finding is in line with data from cross-sectional studies in children and adolescents with obesity in European German-speaking countries.[9,10] Other cross-sectional studies also showed that higher degree of obesity in children was associated with higher risk for metabolic derangements.[27,28] Given the longitudinal association, the present study provides a deeper insight of causal association between degree of obesity and metabolic complications. Compared to class 1 obesity, severe obesity might contribute to a higher overload in hepatic fat accumulation and dysregulation of insulin signalling, subsequently.[29] Hepatocyte fat accumulation and insulin resistance have long been suggested as the "first hits" on the pathogenesis of NAFLD.[29] Furthermore, both hepatic and peripheral insulin resistance promotes proinflammatory cytokines,[28,29] which may contribute to liver damage.

Some studies in children and adolescents with obesity have shown that IFG is more common in children and adolescents with NAFLD compared with those without NAFLD.[11,12] IFG and NAFLD have been related to similar pathophysiology involving insulin resistance.[29] The present study confirms previous findings and contributes additional evidence that IFG diagnosis seems to precede the diagnosis of NAFLD in children and adolescents with obesity. In the present study, individuals with IFG were five times more likely to have later

NAFLD diagnosis, after controlling for other risk factors. However, since we did not have the real date of onset (if there is such) of IFG and NAFLD, but only time of diagnosis, it is not possible to determine the causality. In addition, given a small number of individuals with IFG in this study, the finding should be interpreted with caution. On the other hand, a study in Israeli adolescents showed that NAFLD in adolescents was a risk factor for type 2 diabetes in young adults.[30] It is therefore possible that the association of NAFLD with impaired glucose metabolism in paediatric population is bidirectional, as it has been shown in an adult study.[31] Nonetheless, a possible practical implication of the current finding is that children and adolescents with obesity and IFG should be closely monitored for NAFLD diagnosis later in life. Future research is needed to address the causal association of IFG in the development of NAFLD.

The association of NAFLD and high triglycerides among children and adolescents with obesity has previously been found in epidemiological studies in Israel[32] and German-speaking countries.[10] The results of our study indicate that NAFLD among children and adolescents with obesity may be anticipated by a high level of triglycerides. Moreover, the findings from the previous epidemiological studies[10,32] were not adjusted, whereas our result was independent of age, sex, degree of obesity and the presence of IFG.

Several non-randomized intervention studies in children and adolescents with obesity and NAFLD have shown that lifestyle intervention inducing weight loss is efficacious in reducing NAFLD prevalence.[14–17] However, the lifestyle interventions and weight measurements in the previous studies were performed for short-term periods, i.e., between 10 weeks and 15 months. A longitudinal study of obesity treatment follow-up in Swedish children and adolescents found that majority of individuals who were non-responders during the first year of treatment had a clinically significant weight loss after 3 years.[33] Hence, since short-term treatment is not the optimal way to forecast long-term treatment outcome, we investigated the effect of obesity treatment outcomes in a real-life setting, with a median treatment duration of 32 months. We found that individuals with relative weight loss of at least 0.25 BMI SDS units reached at least a 44% relative risk reduction of NAFLD. This finding underscores the importance of successful obesity treatment, not only for reversing NAFLD but also for the prevention of NAFLD in children and adolescents with obesity. In addition, the present study adds to the evidence that the previously defined term of clinically relevant weight loss, defined as a BMI SDS reduction of at least 0.25 unit, does not only improve cardiovascular risk factors[24] but also is a protective factor for NAFLD among children and adolescents with obesity.

The present study also showed that an increase in BMI SDS was associated with a greater future risk of NAFLD. The risk of NAFLD seemed to increase exponentially with increase in BMI SDS gain, irrespective of degree of obesity between 1 and 6 years prior to index date. A large prospective cohort study among school-aged children in Denmark showed that BMI gain in childhood, independent of initial BMI, was associated with adult NAFLD with median age of NAFLD diagnosis at 55 years.[34] Taken our finding and the finding from the Danish study together, it is reasonable that weight gain in children and adolescents with obesity contribute to early development of NAFLD.

The present study showed a negative effect of IFG and elevated triglycerides on NAFLD, but this effect became insignificant when a clinically relevant weight loss was achieved. These results suggest that a clinically relevant weight loss may restore glucose metabolism and reverse high level of triglycerides in children and adolescents with obesity. However, these results should be interpreted with caution, due to limited power.

It has previously been reported that paediatric NAFLD is more common in certain subpopulations, such as Caucasian, Asian and Hispanic.[1] In the present study, we found a null association between Nordic background and NAFLD. On the other hand, a study in adults in Sweden demonstrated NAFLD prevalence was higher in individuals born in Middle East countries compared to individuals born in Sweden.[35] The null association of Nordic background with NAFLD in this study may be due to the categorization of ethnic background or lack of statistical power to detect the difference. Moreover, we cannot exclude that ethnicity and/or genetic components may differ in cases which are more prone to develop NAFLD.

4.1 | Limitations and strengths of the study

Several strengths of this study need to be acknowledged. Firstly, the primary strength of the study is the data taken from the national registers that covered NAFLD diagnosis both in inpatient and outpatient specialized care from across the nation. This strengthens the external validity of the study. Secondly, unlike previous studies investigating the associations of NAFLD with metabolic parameters in children and adolescents with obesity,[10–13,25,26] we assessed such associations longitudinally, making it possible to speculate in the direction of causality. Thirdly, although paediatric fatty liver disease encompasses a heterogenous group of disorders,[1] we have been able to exclude systemic disorders that may cause fatty liver disease. Finally, as all children in Sweden have free access to healthcare, the risk for selection bias due to a financial situation is likely to be low.

Although simple workup such as serum ALT has been considered as an indirect marker of liver steatosis in children with obesity, liver biopsy remains the gold standard of NAFLD diagnosis.[1,2] The main limitation of the present study is the lack of liver biopsy data of all individuals included in the study, while it is useful to distinguish histological patterns of paediatric NAFLD. Despite the lack of biopsy data, we showed that NAFLD cases in this study were diagnosed in paediatric or gastroentero-hepatology care setting. Serum ALT was not included as a main exposure in the present study given that serum ALT is usually used to support a diagnosis of NAFLD rather than be considered as a potential risk factor for NAFLD. Nevertheless, we report that elevated ALT more than two times normal shortly prior to NAFLD diagnosis was more prevalent among cases compared to controls. Another limitation is that physical examination data, including clinical presentations of insulin resistance (such as acanthosis nigricans and hirsutism due to hyperandrogenism), was not available in the present study. In addition, the generalizability to other populations with different ethnic background may be limited.

Since NAFLD often remains silent and underscreened,[36] individuals in the control group might have undiagnosed NAFLD. Hence, misclassification bias might occur. However, misclassification of the outcome in the present study, if any, is believed to be unrelated to the identified risk factors. Thus, it would not change the direction of the association, although the magnitude of the association may be underestimated.[37] To better reflect fatty liver in children, a term change from NAFLD to "metabolic associated fatty liver disease" (MAFLD) has been proposed.[38] However, the new term MAFLD has also been questioned.[39,40] For instance, some inherited diseases with manifestation of liver steatosis (e.g., Wilson disease, disorders of carbohydrate metabolism) have also a metabolic origin.[39,40] Further studies are needed to assess the clinical and public health implications of changing term NAFLD to MAFLD, more importantly in children with obesity.

5 | CONCLUSIONS

Severe obesity, IFG and high triglycerides are risk factors for NAFLD in children and adolescents with obesity. Yet, clinically relevant weight loss almost eliminates the risk for future NAFLD, regardless of other risk factors. Our findings emphasize the importance of NAFLD screening in children and adolescents with severe obesity, IFG and high triglycerides, as well as the importance of weight loss as prevention of NAFLD.

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CONFLICT OF INTEREST

No conflict of interest was declared.

AUTHOR CONTRIBUTIONS

Emilia Hagman and Resthie R. Putri conceptualized the study, carried out the data management and statistical analysis. Emilia Hagman performed the register linkage and project management. All authors were involved in the interpretation of the results. Resthie R. Putri drafted the manuscript, and all other authors edited the manuscript. All authors approved the final version for submission and agree to be responsible for its contents.

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SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

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