INTRODUCTION

Anaemia is one of the most prevalent public health problems, and it affects more than a quarter of the global population. The highest prevalence is in preschool children and menstruating adolescent girls and women.1 Iron deficiency is the greatest cause of anaemia in children and adolescents, but the prevalence of iron deficiency and anaemia varies greatly worldwide.1-6 These variations may be related to the diversity of the study populations, as well as to different definitions of both iron deficiency and anaemia. Although adolescents are described as a risk group for developing iron deficiency and iron deficiency anaemia,1,2 a limited number of studies from Western countries have described the magnitude of anaemia and iron deficiency. In addition, little is known about the incidence and chronicity of both iron deficiency and anaemia in this age group.7

The causes of iron deficiency and iron deficiency anaemia are multifactorial.2,8 Infants and children are at greater risk of iron deficiency as rapid growth means that they have high iron requirements.9

Abstract

Aim: The combination of iron deficiency and anaemia is a major health problem, and adolescents are an at-risk group. The main aim of this study was to explore the magnitude of these conditions among adolescents aged 15-19 and identify possible associated risk factors.

Methods: This population-based longitudinal study of adolescents in North Norway was conducted in 2010-2011, with a follow-up two years later. Repeated measurements of iron deficiency and anaemia and its possible risk factors were studied in 309 girls and 273 boys.

Results: Iron deficiency and anaemia were found in 18.1% and 19.9% of girls and 1.6% and 2.9% of boys in the first study and about half of the cases were chronic two years later. Most girls had moderate iron deficiency (14.5%) and mild anaemia (16.0%). Daily milk consumption was associated with increased iron deficiency in girls (odds ratio 2.3, 95% confidence interval 1.1–4.9), and the most physically active girls had the lowest levels of iron deficiency (odds ratio 0.4, 95% confidence interval 0.2–0.9). Iron deficiency was the most important risk factor for chronic anaemia in girls.

Conclusion: The results of this study highlight the importance of iron deficiency screening and treatment for adolescent girls.

KEYWORDS
anaemia, epidemiology, iron deficiency, nutritional risk factors, physical activity
Adolescents are also vulnerable to developing iron deficiency and iron deficiency anaemia, especially adolescent girls who experience increased iron losses due to the onset of menstruation. Iron deficiency in children and adolescents can also be caused by low levels of iron in their diet.

The main aim of this study was to describe the prevalence, incidence, chronicity and magnitude of both iron deficiency and anaemia among adolescents in the general population. We also wanted to analyse possible risk factors associated with iron deficiency and anaemia. The main cause of anaemia was presumed to be iron deficiency in our study cohort. The relationships between iron deficiency and different anaemia definitions were included in our analyses for the same reason.

2 | METHODS

This study was conducted as a part of the Tromsø Study, which is a longitudinal population-based study carried out in the municipality of Tromsø in Northern Norway. Tromsø is the ninth most populous municipality in Norway, with a population of more than 70,000 people. In 2010–2011, the Tromsø study was expanded to recruit adolescents from the first Fit Futures study, including all students aged 15–17 years who were in their first year of high school. The participants completed a health questionnaire and participated in physical measurements and medical examinations, including blood sampling. The same students were invited to take part in a second survey two years later, in 2012–2013, when they were 17–19 years of age and in their last year of high school (Figure 1). Haemoglobin and the iron status measurements from both study waves were included in the present study, in addition to data from the health questionnaire. Most of the participants were born in Norway (95.4%), 3.8% were of indigenous Sami ethnicity and 1.3% were of African or Asian ethnic origin.

Both studies were approved by the Regional Committee for Medical and Health Research Ethics in North Norway. Written informed consent was obtained from the participants before they were included in the study.

3 | MEASUREMENTS

3.1 | Anaemia, iron deficiency and iron deficiency anaemia

The World Health Organization (WHO) definitions of anaemia were used, with a cut-off haemoglobin level of <12 g/dl for the girls and of <13 g/dl for the boys. The anaemic cases were further subdivided into mild or moderate and severe anaemia, as shown in Table 2. Iron deficiency was defined by a serum ferritin (SF) level of below 12 µg/L, as suggested by the WHO, based on epidemiological studies of iron deficiency. The degree of iron deficiency was based on SF measurements: severe (<5 µg/L), moderate (5-<12 µg/L) and mild (12-16 µg/L). Iron deficiency anaemia was defined as the combination of anaemia and iron deficiency. Chronic cases were defined as repeated measurements of both anaemia and iron deficiency at baseline and in the second study two years later. Participants with iron deficiency in the first study were informed of their abnormal blood values and advised to visit their family doctor. No public screening programmes for iron deficiency were available in Norway, or the specific geographical area we were looking at, during the two study periods.

Participants with a self-reported disease, chronic inflammation, risk of malabsorption, and those who used iron supplements, were excluded from the statistical analyses, so that we had a healthy reference population sample. Individuals with C-reactive protein above 5 mg/dl were also excluded from the SF analyses. An alternative anaemia definition was used in the study analyses. This was below the 2.5th percentile of the haemoglobin measurements among the participants with a sufficient iron store (SF >16 µg/L) and mean corpuscular erythrocyte volume of >83 f/L.

3.2 | Possible risk factors for iron deficiency and anaemia

Possible risk factors for iron deficiency and anaemia were only studied for the girls, due to the low prevalence of iron deficiency and anaemia in boys. The analyses of possible iron deficiency and anaemia risk factors included the girls’ age, their questionnaire answers and their BMI from the first study. Questions on food habits were adapted from the Food Frequency Questionnaire. The regularity of main meals and how often the subjects consumed fruit, vegetables and a glass of milk were subdivided into daily or less frequent intake. The reported number of days and hours they were physically active per week were dichotomised into at least four days and four hours per week or less to measure their physical activity levels. Questions on menstruation included the age at onset, divided into less than 12, 12–13 and 14 or more years, and the number of days between each menstruation cycle, namely less than 27 days or 27...
or more days. The iron status in the baseline study was included to identify the risk of anaemia. We also used data from the first study to identify the risk for chronic iron deficiency and chronic anaemia in the second study two years later.

### 3.3 Statistical analyses

SPSS version 26 (IBM Corp) was used to analyse the study data. The Student’s *t* test was used to study normally distributed data, the Mann-Whitney *U* test was used for non-parametric distributed data and the chi-square test for categorical data. The repeated study measurements were analysed by the paired sample *t* test for the normally distributed data, the related-samples Wilcoxon signed rank test for non-parametric repeated data and the related-samples McNemar test for repeated categorical measurements. Logistic regression was used to identify possible associations and risk factors for both iron deficiency and anaemia in both studies, and the results were reported as odds ratios (OR) and 95% confidence intervals (95% CI).

### 4 RESULTS

The first study comprised 871 students aged 15–17 (53.2% boys), and the second study comprised 608 students aged 17–19 years (46.1% boys). There were 582 students (46.9% boys) who completed the study at both ages (Figure 1). One-third of the participants in the first study reported one or more chronic medical conditions, and asthma and allergic diseases were the most common. Coeliac disease was reported by five participants and two reported recognised chronic anaemia. There were three participants who reported regularly taking oral iron supplements and 22.0% reported taking vitamin and mineral supplements, but without detailed information about whether they contained iron. More than a third (36.0%) of the girls reported that they regularly used oral contraceptives.
Repeated measurements of haemoglobin and iron status

4.1 Repeated measurements of haemoglobin and iron status

Both the girls and boys showed a significant increase in SF level between the two study periods (Table 1). The SF level at the 2.5th percentile was significantly lower in girls than the WHO SF cut-off value for iron deficiency definition in both studies (p < 0.01).

The haemoglobin level was stable in girls, but increased significantly in boys, with a mean difference of Hgb 0.22 g/dl (p < 0.01). The lower haemoglobin percentile in the reference and the iron sufficient population corresponded well with the WHO cut-off level for defining anaemia in boys (Hgb < 13 g/dl). When it came to the girls, the same 2.5th percentiles of haemoglobin concentrations (Hgb = 11.4 g/dL) were significantly lower than the WHO haemoglobin cut-off level for the definition of anaemia (Hgb < 12 g/dL) in both studies (p < 0.01).

4.2 The prevalence and incidence of iron deficiency, anaemia and iron deficiency anaemia

Table 2 shows that one-fifth (19.9%) of the adolescent girls had anaemia in the first study, according to the WHO definition. The prevalence of anaemia and iron deficiency in girls was seven and 11 times higher than in boys. For both girls and boys, the prevalence of iron deficiency, anaemia and iron deficiency anaemia were lower in the second study than the first (Table 2). Most of the anaemic girls had mild anaemia in both studies (80.4% in the first and 77.4% in the second study), as did all the anaemic boys. The decrease in iron deficiency over the two-year period was significant in girls and the same was seen in the mild iron deficiency cases in boys. Both the incidence and chronicity of the iron deficiency cases in boys were relatively low, and they were significantly lower than in the girls (p < 0.01).

Approximately half (48.2%) of the anaemic girls and one-fifth (20.7%) of the anaemic boys had iron deficiency anaemia in the first study, according to the WHO anaemia definitions. When we used the lower haemoglobin cut-off of the 2.5th percentile in the iron sufficient reference population, namely a haemoglobin of <11.4 g/dL, this identified iron deficiency anaemia in two-thirds of the girls. This provided a significantly higher positive predictive value compared with the WHO cut-off value of 12 g/dL, which only identified half of the iron deficiency anaemia cases. The negative predictive value for iron deficiency anaemia remained relatively high and stable for both cut-off values (Table 2).

4.3 Risk factors of iron deficiency and anaemia among girls

Iron status was the most important predictor of anaemia for girls in the first study and also for chronic iron deficiency and chronic anaemia in the second study two years later. The associations were significantly related to the severity of iron deficiency (Table 3). We found that 73.0% of the girls with chronic anaemia in the baseline study had iron deficiency in the second study.

| TABLE 1 | Measurements of haemoglobin concentration and serum ferritin in the two studies |
| --- | --- | --- | --- |
| **First study (2010–2011) (n = 871)** | **Second study (2012–2013) (n = 608)** |
| Total study population | Reference population | Percentiles reference population | Total study population | Reference population | Percentiles reference population |
| **Girls** | **Haemoglobin, g/dL** | 401 | 12.6 (1.0) | **2,5** | 12.7 (0.9) | **97.5** | 14.2 |
| | n | Mean (SD) | n | Mean (SD) | 2.5 | 97.5 |
| | **Haemoglobin, g/dL** | 391 | 12.7 (0.9) | 10.9 | 14.2 |
| | **Serum ferritin, µg/L** | 235 | 12.9 (0.8) | 11.4 | 14.5 |
| | 408 | 30.2 (21.8) | 376 | 28.5 (19.0) | 4.0 | 77.6 |
| **Boys** | **Haemoglobin, g/dL** | 455 | 14.6 (0.9) | **2,5** | 14.6 (0.8) | **97.5** | 16.2 |
| | n | Mean (SD) | n | Mean (SD) | 2.5 | 97.5 |
| | **Haemoglobin, g/dL** | 442 | 14.6 (0.8) | 13.0 | 16.2 |
| | **Serum ferritin, µg/L** | 349 | 14.6 (0.8) | 13.0 | 16.1 |
| | 463 | 55.2 (34.1) | 410 | 51.9 (27.0) | 13.0 | 113.7 |
| **n** | **Mean (SD)** | **n** | **Mean (SD)** | **n** | **Mean (SD)** | **n** | **Mean (SD)** |
| **Girls** | **Haemoglobin, g/dL** | 322 | 12.7 (0.9) | **2,5** | 12.9 (0.8) | **97.5** | 14.5 |
| | n | Mean (SD) | n | Mean (SD) | 2.5 | 97.5 |
| | **Haemoglobin, g/dL** | 317 | 12.7 (0.9) | 10.7 | 14.5 |
| | **Serum ferritin, µg/L** | 287 | 39.4 (28.9)** | 6.0 | 115.4 |
| **Boys** | **Haemoglobin, g/dL** | 279 | 14.8 (0.9)** | **2,5** | 14.8 (0.9)** | **97.5** | 16.2 |
| | n | Mean (SD) | n | Mean (SD) | 2.5 | 97.5 |
| | **Haemoglobin, g/dL** | 273 | 14.8 (0.9)** | 13.1 | 16.2 |
| | **Serum ferritin, µg/L** | 227 | 14.8 (0.9)** | 13.1 | 16.2 |
| *p < 0.05 and **p < 0.01 in the analyses of changes of repeated measurements in the first and second study (paired sample t tests). |
| aReference population excluded participants with chronic diseases and measurement outliers (Chauvenet’s criterion). |
| bIron sufficient reference population with SF >16 µg/L and mean corpuscular erythrocyte volume >83 fL.
TABLE 2  The prevalence and incidence of iron deficiency, anaemia and iron deficiency anaemia in both studies

|                      | First study  | Second study | Incidence %/year | Chronic % |
|----------------------|--------------|--------------|------------------|-----------|
|                      | (408 girls/463 boys) | (328 girls/280 boys) |                  |           |
| Iron deficiency      |              |              |                  |           |
| Girls                |              |              |                  |           |
| SF <12 µg/L          | 18.1         | 12.4 *       | 2.6             | 71        |
| Mild (SF <16 and ≥12 µg/L) | 9.6         | 7.2          |                  |           |
| Moderate (SF <12 and ≥5 µg/L) | 14.5       | 11.7 *       |                  |           |
| Severe (SF <5 µg/L)  | 3.6          | 0.7 **       |                  |           |
| Boys                 |              |              |                  |           |
| SF <12 µg/L          | 1.6          | 0.8          | 0.15             | 0.5       |
| Mild (SF <16 and ≥12 µg/L) | 3.5          | 0.8 **       |                  |           |
| Moderate (SF <12 and ≥5 µg/L) | 1.6         | 0.8          |                  |           |
| Severe (SF <5 µg/L)  | 0            | 0            |                  |           |
| Anaemia              |              |              |                  |           |
| Girls                |              |              |                  |           |
| Hgb <12 g/dl         | 19.9         | 16.4         | 3.2              | 10.0      |
| Hgb <11.4 g/dL       | 4.8          | 5.3          | 1.1              | 2.4       |
| Mild Hgb <12 and ≥11 g/dl | 16.0      | 12.7         |                  |           |
| Moderate Hgb <11 and ≥8 g/dl | 3.7         | 3.4          |                  |           |
| Severe Hgb <8 g/dl   | 0.2          | 0.3          |                  |           |
| Boys                 |              |              |                  |           |
| Hgb <13 g/dl         | 2.9          | 1.8          | 0.8              | 0.4       |
| Iron deficiency anaemia |            |              |                  |           |
| Girls                |              |              |                  |           |
| Hgb <12 g/dL and SF <12 µg/L | 9.6        | 53.6% / 89.4% | 6.6              | 38.0% / 92.7% |
| Hgb <11.4 g/dL and SF <12 µg/L | 3.2        | 66.7% / 86.0% | 3.5              | 66.7% / 90.7% |
| Boys                 |              |              |                  |           |
| Hgb <13 g/dL and SF <12 µg/L | 0.6      | 0.4          | 0.0              | 0.4       |

* p < 0.05 and ** p < 0.01 in the analyses of prevalence changes by the Related-Samples McNemar test. Cases with C-reactive protein >5 mg/L were excluded from the analyses of iron deficiency. PPV/NPV = iron deficiency positive predictive values and negative predictive values of anaemia in girls by the two different anaemia definitions: (1) WHO cut-off (Hgb <12 g/dl) and (2) below the lower haemoglobin limit at the 2.5th percentile in the iron sufficient reference population (Hgb < 11.4 g/dl).
### TABLE 3  Association of iron deficiency, chronic iron deficiency and chronic anemia in girls

| Iron deficiency at baseline | Odds ratio (95% CI) | 
|----------------------------|---------------------|
|                            | Anaemia Hgb <12 g/dl | Chronic iron deficiency SF <12 µg/L | Chronic anaemia Hgb <12 g/dl |
| First study                |                     |                                 |
| Serum ferritin (SF) >16 µg/l (n = 293) | 1.0 | 1.0 | 1.0 |
| Mild (SF 12–16 µg/l) (n = 38) | 2.0 (0.8–4.6) | 7.3 (2.5–21.5) ** | 2.3 (0.7–7.7) |
| Moderate (SF 5–12 µg/l) (n = 56) | 5.5 (2.9–10.5) ** | 10.8 (4.2–27.4) ** | 3.0 (1.1–8.6) * |
| Severe (SF <5 µg/l) (n = 14) | 95.8 (12.2–755.1) ** | 72.0 (16.6–312.2) ** | 70.0 (7.9–620.3) ** |

*p < 0.05 and **p < 0.01 based on simple logistic regression of iron deficiency subgroups (reference = SF>16 µmol/l) and anaemia in the first study and chronic anaemia and iron deficiency in the second study.

Drinking one or more glasses of milk per day at the time of the first study was significantly associated with iron deficiency when we compared those subjects with the girls who drank less than that (OR 2.0, 95% CI 1.1–3.4). The regularity of main meals and how frequency participants ate fruit and vegetables were not associated with iron deficiency.

The 15-year-old girls in the present study had a significantly higher risk of iron deficiency than the 16- and 17-year-old girls in the first study (OR 4.5, 95% CI 1.3–15.7). BMI was not a significant predictor of iron deficiency or anaemia. We found no association between the age when menstruation started and iron deficiency in the first study. The number of days between each menstruation cycle and the use of oral contraceptives were not significantly associated with iron deficiency or anaemia. Finally, the most physically active girls had a lower risk of iron deficiency than the other female participants (OR 0.4, 95% CI 0.2–0.9).

Only daily consumption of one or more glasses of milk in the first study was positively associated with chronic iron deficiency in the second study two years later (OR 2.3 with 95% CI 1.1–4.9). The remaining lifestyle factors we have described were not associated with chronic iron deficiency.

### 5 | DISCUSSION

This study found a high prevalence of iron deficiency and anaemia in a large representative population of adolescent girls, with a lower prevalence in boys. These findings agreed with several previous studies. Despite the high prevalence of iron deficiency that was observed, most of the girls, and all of the boys, only had mild to moderate iron deficiency and mild to moderate anaemia. That was in accordance with the few earlier studies that were available, including similar results on the severity of iron deficiency and anaemia. About half of the iron deficiency cases in the first study had chronic iron deficiency in the second study. Furthermore, the annual incidence of iron deficiency was found to be relatively low. We believe that these results are important, due to the lack of previous and comparable longitudinally designed epidemiological studies. As expected, iron deficiency was the most common cause of anaemia in girls. Importantly, it was the also the most significant risk factor for both chronic iron deficiency and anaemia and these results were strengthened by clear associations with the severity of their iron deficiency.

Mean iron stores increased between the first and second studies, while the prevalence of iron deficiency decreased in both girls and boys. Samuelson et al. published one of the few comparable longitudinal studies in adolescents aged 15–17 and described a similar increase in the boys’ iron stores and stable rates among the girls. Our results were also comparable with several previous cross-sectional, population-based studies of different age-groups, which showed that iron stores increased in both genders. Other studies showed that high iron deficiency was prevalent among the youngest teenage girls. Ferrari et al. carried out a study on the prevalence of iron deficiency in adolescent girls aged 12–17 years in 10 European countries. They reported that 21.0% had iron deficiency (SF < 15 µg/L), and those findings were somewhat lower than the 27.7% in the present study when a slightly higher cut-off point was used (SF < 16 µg/L). In contrast, the figures for the boys of the same age were 13.8%, which was significantly higher than the 5.1% in the present study. The overall discrepancies between the prevalence of iron deficiency in various studies may be explained by the different ages of the study participants, their country and ethnicity and the various definitions of iron deficiency.

Growing children and adolescents have higher iron requirements than adults. Insufficient dietary intake of iron is one of the main causes of iron deficiency. Intake can also be influenced by dietary factors that inhibit intestinal iron absorption, such as calcium, or facilitate it, such as vitamin C. The first study showed a significant association between girls drinking one or more glasses of milk and iron deficiency, and this level of consumption was also associated with chronic iron deficiency in the second study two years later. Studies had previously shown an association between milk consumption and iron deficiency in younger children, but we were unable to find similar results in adolescent studies. Therefore, this study fills a gap in our knowledge of the links between milk consumption in adolescent girls and iron deficiency. In contrast, infrequent intake of both fruit and vegetables was not associated with iron deficiency, which had previously been identified as risk factors. It was not
possible to study other well described dietary risk factors for iron deficiency, such as insufficient consumption of meat and grain products and a vegetarian diet, as these were not included in the study questionnaire.

It has been reported that athletes have high iron requirements and a high risk of iron deficiency. In contrast, the most physically active girls in our study had a significantly lower risk of iron deficiency than those who were less active. This could be explained by their healthy lifestyle, including diets with sufficient amounts of iron and a more moderate level of exercise than the athletes in that study. The girls’ body mass index was not associated with iron deficiency in our subjects, in contrast to previous reports. We did not include any possible associations between iron deficiency and growth spurts and changes in body mass index during puberty in our study. This was due the lack of repeated anthropometric measures in the second study.

Menstrual blood loss is known to be the most important risk factor for iron deficiency and anaemia in adolescent girls. Questions about the degree of menstrual blood loss, and possible hypermenorrhoea, were not included in our questionnaire, which is clearly an important study limitation. Another study limitation was that information was not requested on the use of iron supplements before and during the second study wave. However, participants with iron deficiency in the first study were advised to contact their family doctor to follow this up. We found no association between oral contraceptives and iron deficiency, which is often used to reduce menstrual losses in girls with hypermenorrhoea.

All the risk factors for iron deficiency that were described in our study were significant risk factors for anaemia, but the same associated risk factors for anaemia were not independent of iron deficiency. These results confirm that iron deficiency is the most important cause of anaemia in the western world. The results also showed what we expected, as we studied a homogenous study population, with mainly healthy adolescents and minimal ethnic variations. Those factors lowered the risk of haemoglobinopathies and other causes of anaemia. Despite this, only about half of the girls who met the WHO definition for anaemia also had iron deficiency at the sometime. However, this increased to two-thirds of the anaemic cases with haemoglobin concentrations below 11.4 g/dl. This cut-off point corresponded to the lower haemoglobin limit of the 2.5th percentile in the iron sufficient reference population in the study. The latter haemoglobin cut-off point for defining anaemia in girls seemed to be more adequate. It also agreed with some previous population-based studies that included reference data on the haemoglobin distribution and the lower limits in age-matched adolescent girls.

6 | CONCLUSION

Mild-to-moderate iron deficiency and anaemia were highly prevalent in the adolescent girls in our large longitudinal population-based study and about half of the cases were chronic, as they appeared at both study ages. Iron deficiency was the most important risk factor for anaemia, which underlines the importance of screening for iron deficiency in clinical practice. It also indicates the need for public iron deficiency screening and treatment programmes for adolescent girls.

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CONFLICTS OF INTEREST

The authors have no conflicts of interest to declare.

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