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# Reproductive epidemiology

# Growth of singletons born after frozen embryo transfer until early adulthood: a Finnish register study

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## ABSTRACT

**STUDY QUESTION:** Are there growth differences between singleton children born after frozen embryo transfer (FET), fresh embryo transfer (ET), and natural conception (NC)?

**SUMMARY ANSWER:** Adolescent boys born after FET have a higher mean proportion and increased odds of overweight compared to those born after fresh ET.

**WHAT IS KNOWN ALREADY:** Children born after FET have higher mean birthweights and an increased risk of large-for-gestational-age compared to those born after fresh ET and even NC. This raises questions about possible growth differences later in childhood. Previous studies on child growth after FET report partly conflicting results and lack long-term data until adolescence.

**STUDY DESIGN, SIZE, DURATION:** This was a cohort study based on national population-based registers, the Finnish Medical Birth Register and the Register of Primary Health Care visits, including singletons born after FET (n = 1825), fresh ET (n = 2933), and NC (n = 31 136) in Finland between the years 1995 and 2006.

**PARTICIPANTS/MATERIALS, SETTING, METHODS:** The proportions of overweight (i.e. age- and sex-adjusted ISO-BMI for children ≥ 25) were compared between the groups. Odds ratios (ORs) and adjusted odds ratios (aORs) of overweight were calculated. Adjustments were made for birth year, preterm birth, maternal age, parity, and socioeconomic status. Mean heights, weights, and BMIs were compared between the groups each year between the ages of 7 and 18.

**MAIN RESULTS AND THE ROLE OF CHANCE:** FET boys had a higher mean proportion of overweight (28%) compared to fresh ET (22%, P < 0.001) and NC (26%, P = 0.014) boys. For all ages combined, the aOR of overweight was increased (1.14, 95% CI 1.02–1.27) for FET boys compared to fresh ET boys. For girls, the mean proportions of overweight were 18%, 19%, and 22% for those born after FET, fresh ET, and NC, respectively (P = 0.169 for FET vs fresh ET, P < 0.001 for FET vs NC). For all ages combined, FET girls had a decreased aOR of overweight (0.89, 95% CI 0.80–0.99) compared to NC girls. Growth measurements were available for 6.9% to 30.6% of FET boys and for 4.7% to 29.4% of FET girls at different ages.

**LIMITATIONS, REASONS FOR CAUTION:** Unfortunately, we were not able to adjust for parental anthropometric characteristics. The growth data were not available for the whole cohort, and the proportion of children with available measurements was limited at the start and end of the follow-up. During the study period, mainly cleavage stage embryos were transferred, and slow freezing was used for ART.

**WIDER IMPLICATIONS OF THE FINDINGS:** The risk of overweight among FET boys warrants further research. Future studies should aim to investigate the mechanisms that explain this sex-specific finding and combine growth data with long-term health data to explore the possible risks of overweight and cardiometabolic disease in adulthood.

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**Keywords:** ART / FET / frozen embryo transfer / child growth / long-term outcome

## Introduction

The growth of children born after frozen embryo transfer (FET) has been of interest since their increased risks of high birth weight and large-for-gestational-age (LGA) were first documented more than 10 years ago, and subsequently verified in large cohorts and meta-analyses (Pelkonen et al., 2010; Pinborg et al., 2014; Maheshwari et al., 2018; Sha et al., 2018; Terho et al., 2021a). LGA and high birthweight in general population have been shown to be associated with increased risks of obesity, diabetes, cardiovascular disease, and certain malignancies later in life (Magnusson et al., 2021; Scifres, 2021).

Previous studies on growth after FET have presented partly contradictory results. Some have reported no differences in growth until the age of 5 (Ainsworth et al., 2019; Turner et al., 2020; Terho et al., 2021b). In contrast, a British study reported FET children to be taller and fresh embryo transfer (ET) children to have a smaller BMI at the age of 4 to 7 compared to naturally conceived (NC) children (Hann et al., 2018). Previous literature has included only a limited number of FET children in the advanced adolescent years, reporting no significant differences in growth (Magnus et al., 2021; Elhakeem et al., 2022).

In Europe, 3.5% of children are born after ARTs. The use of FET is on the rise, comprising 35.5% of all treatment cycles in 2018, and even freeze-all cycles are implemented widely (Wyns et al., 2022). Therefore, it is of utmost importance to continue providing data on the long-term safety of FET. The aim of this study was to investigate the growth of singleton children born after FET compared to fresh ET and NC until early adulthood in the setting of reliable Finnish health registers.

## Materials and methods
### Study population
This register-based cohort study included live-born singletons born between the years 1995 and 2006 after FET (n = 1825) and fresh ET (n = 2933). Women who underwent ART treatment leading to birth were first identified in patient registers of the infertility clinics of the University Hospitals and the Väestöliitto Fertility Clinics in Oulu and Helsinki. ETs during the study period were mainly performed at the cleavage stage and mainly slow freezing was used. By using the personal identification numbers of the women, the corresponding births were matched with data from the Finnish Medical Birth Register (FMBR).

A 10% random sample of NC controls (n = 31 136) from the same time period, matched for area of residence, was obtained from the FMBR. Previous publications on this cohort include papers covering the perinatal health (Pelkonen et al., 2010), major congenital anomalies (Pelkonen et al., 2014), somatic health of children until the age of 3 (Pelkonen et al., 2015), and their health until early adulthood (Terho et al., 2022). For the present study, data from the FMBR were combined with data from the Register of Primary Health Care visits.

The Finnish Institute for Health and Welfare (THL) is responsible for the FMBR and the Register of Primary Health Care visits. The FMBR includes data on live births and stillbirths with a birth weight of at least 500 g or gestational age of at least 22 weeks, as well as data on the mothers. Data sources of FMBR include maternity hospitals and neonatal units, the Finnish Central Population Register, kept by Digital and Population Data Services Agency, and the Cause-of-Death Register, kept by Statistics Finland.

The Register of Primary Health Care visits covers all outpatient visits in municipal health centers, including school

healthcare, since 2011. The care notifications include data on the personal identity number of the patient, reason for seeking care, diagnoses, procedures, and interventions (THL, 2021). Growth measurement data derived from school health care are included for a portion of Finnish children, where the local electronic patient record system made it possible to transfer growth data to the Register. School health care is a mandatory health service provided by Finnish municipalities to monitor the health, growth, and social wellbeing of students and their families (THL, 2022). Annual check-ups, including growth monitoring, are offered. If growth disturbances, for example overweight, are detected, additional measurements are programmed. We used the information from the first measurement for each age to avoid any bias related to multiple measurements. For this study, we used growth data between ages 7 and 18. The growth data are not necessarily derived from the same children at different ages but enable the comparison of the mean measurements between the study groups.

Data linkage between registers is possible using the unique personal identity numbers assigned to each Finnish citizen immediately at birth or upon immigration. The personal identity numbers are extensively used throughout the society: in healthcare, social welfare system, school system, banking etc. All data were pseudonymized after the register linkage before analyses.

### Study permission and ethical approval
Study permission was granted by Findata, the Social and Health Data Permit Authority (THL/1162/14.05.00/2022 findata-rems-2022/149). Separate ethical approval was not needed since the study is based on register data and no registered person was contacted.

### Outcomes and statistical methods
The International Obesity Task Force (IOTF) age- and sex-adjusted cutoff values for overweight in children (Cole et al., 2000) were used to compare the proportions of overweight (ISO-BMI ≥25) between the FET, fresh ET, and NC groups. Odds ratios (ORs) and adjusted odds ratios (aORs) of overweight were calculated. Adjustments were made for birth year, preterm birth (PTB, <37 gestational weeks), maternal age, parity, and socioeconomic status based on mother's occupation at childbirth (SES). Mean weights (as kg), heights (as cm), and BMIs (defined as weight in kilograms divided by the square of height in meters, $kg/m^2$) were compared between FET, fresh ET, and NC children, yearly between ages 7 and 18. All analyses were performed separately for boys and girls.

Background characteristics were described as mean and SD for continuous variables and as count and percentage for categorical variables. Student's T-test and Chi-square test were used for continuous and categorical variables, respectively. The mean proportions of overweight were compared using the test for relative proportions. Logistic regression was used to analyze the ORs and aORs of children with overweight. Additionally, the odds of overweight were calculated for all age groups combined, adjusting for multiple measurements using generalized estimating equations to avoid bias caused by the number of measurements varying by child (mean 3, range 1–10 per child). A general linear model was used to compare the mean heights, weights, and BMIs among the three study groups.

All statistical analyses were performed using SAS statistical software, version 9.4 (SAS Institute).

## Results

### Background characteristics

Background characteristics of the study population are presented in Table 1. Mothers in the FET group were older (mean 34.3 years) compared to fresh ET (33.9 years, $P = 0.009$) and NC (30.0 years, $P < 0.001$) groups. They were less often primiparous (34.8%) compared to fresh ET (52.3%, $P < 0.001$), but more often compared to NC (31.8%, $P = 0.007$). The number of PTBs in the FET group was lower (6.4%) compared to the fresh ET group (8.5%, $P = 0.008$), but higher compared to the NC group (4.3%, $P < 0.001$). The mean birth weight for the FET group (3554 grams) was significantly higher compared to the fresh ET group (3423 grams, $P < 0.001$) but not compared to the NC group (3544 g, $P = 0.437$). FET boys were significantly more often LGA (4.2%) compared to fresh ET (2.1%, $P = 0.002$) and NC (2.9%, $P = 0.022$) boys. FET girls were significantly more often LGA (3.7%) compared to fresh ET (2.2%, $P = 0.030$) but not compared to NC (2.9%, $P = 0.212$) girls (Table 1).

No statistically significant differences existed between the study groups in the percentage of children with at least one measurement in the Register (FET 55.2%, fresh ET 53.5%, and NC 53.2%; FET vs NC: $P = 0.10$, fresh ET vs NC: $P = 0.72$). The same was true after adjusting for birth year or background factors: maternal age, parity, SES, offspring sex, and PTB (data not shown).

Growth measurements for each age were available for 6.9% to 30.6% of FET boys and for 4.7% to 29.4% of FET girls. For fresh ET, the percentages were 6.5% to 27.2% and 5.9% to 27.3%, and for NC, 5.9% to 25.3% and 5.9% to 26.0% for boys and girls, respectively (Supplementary Tables S1 and S2). The mean and median number of measurements were three per child (range from 1 to 10).

### The proportions and odds of overweight

For boys, the mean proportions of age-specific overweight rates among FET, fresh ET, and NC were 28%, 22%, and 26%, respectively ($P < 0.001$ for FET vs fresh ET, $P = 0.014$ for FET vs NC and $P < 0.001$ for fresh ET vs NC; Fig. 1). In the yearly measurements, FET boys had a significantly increased aOR of overweight in the teen years between ages 14 and 16 compared to fresh ET boys and between ages 15 and 16 when compared to NC boys. For all ages combined, the aOR of overweight was 1.14 (95% CI 1.02–1.27) for FET compared to fresh ET, and 1.08 (95% CI 0.99–1.18) for FET compared to NC (Fig. 2, Supplementary Table S3).

For girls, the mean proportions of overweight among FET, fresh ET, and NC were 18%, 19%, and 22% ($P = 0.169$ for FET vs fresh ET, <0.001 for FET vs NC and $P < 0.001$ for fresh ET vs NC; Fig. 1). FET girls had significantly decreased aORs of overweight between ages 15 and 16 compared to NC. For all ages combined, the aOR of overweight was 0.92 (95% CI 0.81–1.06) for FET

**Table 1.** Background characteristics of the study population.

| Variable | FET n = 1825 | Fresh ET n = 2933 | NC n = 31 136 | P FET vs fresh ET | P FET vs NC | P fresh ET vs NC |
|---|---|---|---|---|---|---|
| Maternal age, y, mean (SD) | 34.3 (4.0) | 33.9 (4.3) | 30.0 (5.4) | **0.009** | **<0.001** | **<0.001** |
| Missing, n (%) | <5 (0.1) | 0 (0) | 71 (0.2) | | | |
| Primiparous, n (%) | 635 (34.8) | 1535 (52.3) | 9893 (31.8) | **<0.001** | **0.007** | **<0.001** |
| Missing (%) | <5 (0.1) | <5 (0.0) | 105 (0.3) | | | |
| Smoking, n (%) | 130 (7.1) | 194 (6.6) | 4707 (15.1) | 0.498 | **<0.001** | **<0.001** |
| Missing, n (%) | 24 (1.3) | 43 (1.5) | 774 (2.5) | | | |
| Maternal SES | | | | 0.629 | **<0.001** | **<0.001** |
| Upper white collar worker, n (%) | 581 (31.8) | 912 (31.1) | 6460 (20.7) | | | |
| Lower white collar worker, n (%) | 798 (43.7) | 1254 (42.8) | 11 873 (38.1) | | | |
| Blue collar worker, n (%) | 166 (9.1) | 286 (9.8) | 3971 (12.8) | | | |
| Other, n (%) | 122 (6.7) | 219 (7.5) | 6150 (19.8) | | | |
| Missing, n (%) | 158 (8.7) | 262 (8.9) | 2682 (8.6) | | | |
| ICSI, n (%) | 536 (29.4) | 965 (32.9) | NA | **0.011** | NA | NA |
| Preterm births, n (%) | 117 (6.4) | 250 (8.5) | 1341 (4.3) | **0.008** | **<0.001** | **<0.001** |
| Extremely preterm, <28 GW, n (%) | 5 (0.3) | 14 (0.5) | 73 (0.2) | 0.279 | 0.736 | **0.013** |
| Caesarean sections, n (%) | 518 (28.4) | 818 (27.9) | 5151 (16.5) | 0.712 | **<0.001** | **<0.001** |
| Missing, n (%) | 0 (0) | 0 (0) | 5 (0.0) | | | |
| Offspring male sex, n (%) | 926 (50.7) | 1500 (51.1) | 15 945 (51.2) | 0.787 | 0.696 | 0.943 |
| Birth weight, All, g, mean (SD) | 3554 (579) | 3423 (594) | 3544 (544) | **<0.001** | 0.437 | **<0.001** |
| Boys, g, mean (SD) | 3614 (587) | 3482 (597) | 3599 (563) | **<0.001** | 0.449 | **<0.001** |
| Girls, g, mean (SD) | 3493 (565) | 3360 (584) | 3486 (518) | **<0.001** | 0.717 | **<0.001** |
| LGA, All, n (%) | 72 (3.9) | 62 (2.1) | 909 (2.9) | **<0.001** | **0.012** | **0.012** |
| Boys, n (%) | 39 (4.2) | 31 (2.1) | 462 (2.9) | **0.002** | **0.022** | 0.063 |
| Girls, n (%) | 33 (3.7) | 31 (2.2) | 447 (2.9) | **0.030** | 0.212 | 0.092 |
| SGA, All, n (%) | 55 (3.0) | 126 (4.3) | 992 (3.2) | **0.025** | 0.683 | **0.001** |
| Boys, n (%) | 26 (2.8) | 51 (3.4) | 517 (3.2) | 0.419 | 0.466 | 0.742 |
| Girls, n (%) | 29 (3.2) | 75 (5.2) | 475 (3.1) | **0.022** | 0.869 | **<0.001** |
| NICU, n (%) | 49 (2.7) | 77 (2.6) | 525 (1.7) | 0.901 | **0.002** | **<0.001** |
| Birth year | | | | **<0.001** | **<0.001** | **<0.001** |
| 1994–1997 | 241 (13.2) | 567 (19.3) | 9937 (31.9) | | | |
| 1998–2000 | 421 (23.1) | 820 (28.0) | 6895 (22.1) | | | |
| 2001–2003 | 533 (29.2) | 773 (26.4) | 6944 (22.3) | | | |
| 2004–2006 | 630 (34.5) | 773 (26.4) | 7289 (23.4) | | | |
| Missing n (%) | 0 (0) | 0 (0) | 71 (0.2)[a] | | | |
| Follow-up time, y, mean (SD)[b] | 18.8 (3.3) | 19.5 (3.6) | 20.5 (4.0) | **<0.001** | **<0.001** | **<0.001** |

[a] Incomplete personal ID code.
[b] Follow-up from birth until 31 December 2020, or death.
Independent samples T-test and Chi-square test.
FET, frozen embryo transfer; ET, embryo transfer; NC, natural conception; y, year; SD, standard deviation; SES, socioeconomic status; ICSI, intracytoplasmic sperm injection; GW, gestational week; g, grams; LGA, large-for-gestational-age; SGA, small-for-gestational-age; NICU, neonatal intensive care unit.
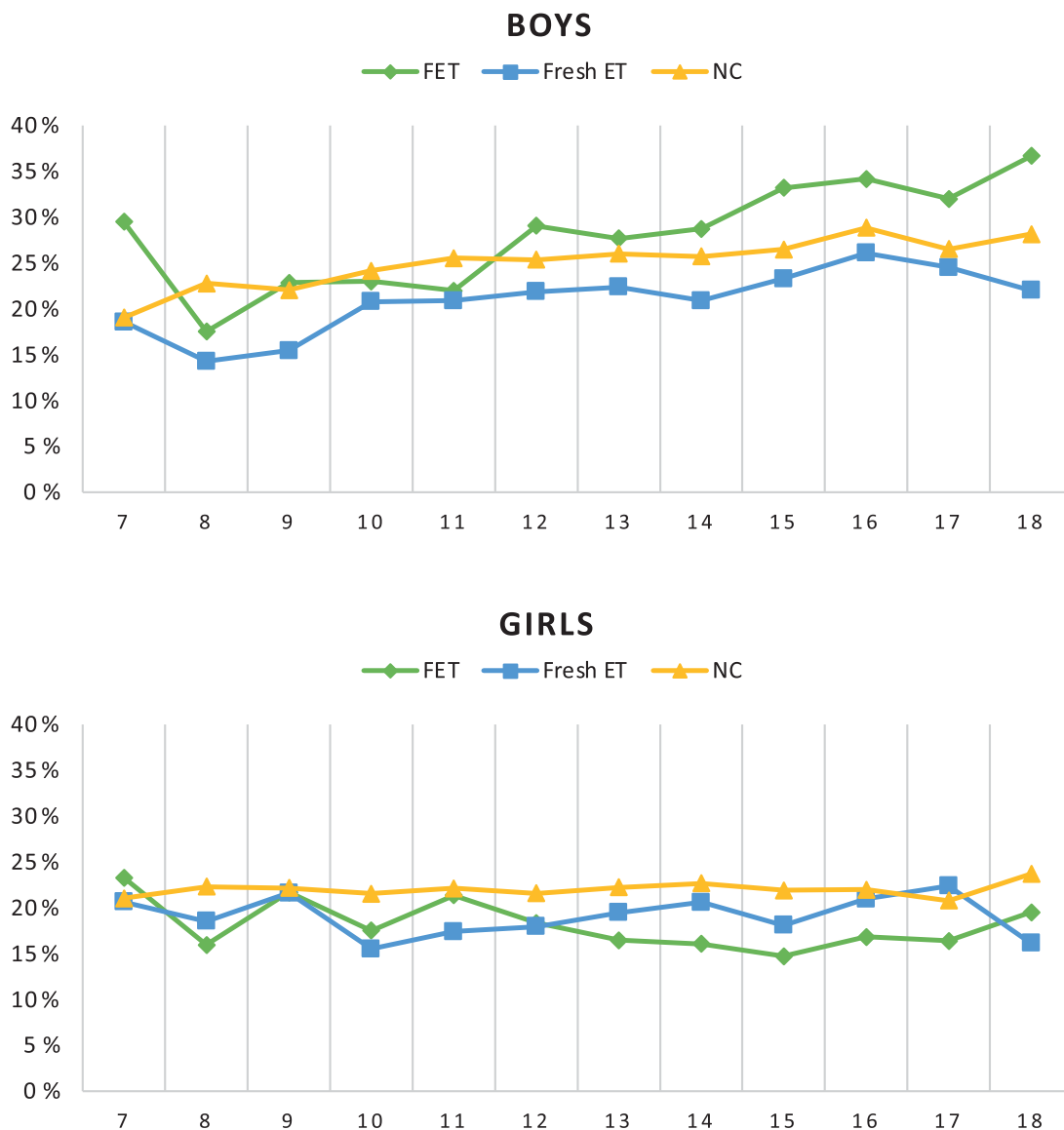Bold indicates statistical significance at $P < 0.05$.

## BOYS



## GIRLS



**Figure 1.** **Proportions of overweight (ISO-BMI ≥ 25) boys and girls by age.** ISO-BMI, the age- and sex-adjusted BMI for children as described by the International Obesity Task Force (IOTF); FET, frozen embryo transfer; ET, embryo transfer; NC, natural conception.

compared to fresh ET, and 0.89 (95% CI 0.80–0.99) for FET compared to NC (Fig. 2, Supplementary Table S3).

### Mean heights, weights, and BMIs

Only sporadic statistically significant changes in height were found between FET and fresh ET. In the crude results on height, FET boys were significantly taller compared to NC boys between ages 9 and 18. Adjustment for birth year, PTB, maternal age, parity, and SES attenuated these differences, although significant differences were still found between ages 11 and 14 (Fig. 3, Supplementary Table S1). FET boys had higher weight and BMI, both crude and adjusted, compared to both fresh ET and NC at several measurements between ages 12 and 18, and 14 and 18, respectively. However, these findings were not consistent for every year of age (Supplementary Table S1). For girls, only sporadic statistically significant differences were found concerning mean heights, weights and BMIs between the groups (Fig. 3, Supplementary Table S2).

## Discussion

The main finding in this register-based study is that towards the end of follow-up, FET boys were heavier with a higher BMI and increased odds of overweight compared to fresh ET. There were no significant differences in height between FET and fresh ET boys. For girls, no significant differences were found for mean height, weight, or BMI between the groups. However, FET girls had slightly decreased odds of overweight compared to NC girls.

There are a few previous studies on the growth of FET children. In our earlier study on a cohort of term-born singletons, we found no differences in the growth of FET children (n = 110), stratified by sex, compared to fresh ET and NC children until the age of 5 (Terho *et al.*, 2021b). In line with this, studies from the USA and the UK found no differences in growth for FET (n = 49 and n = 179, respectively) children until the age of 5 (Ainsworth *et al.*, 2019; Turner *et al.*, 2020). However, another UK cohort study found FET singletons (n = 519) to be taller and fresh ET singletons to have a slightly lower BMI compared to NC singletons at 4 to 7 years of age, after adjusting for offspring sex (Hann *et al.*, 2018).
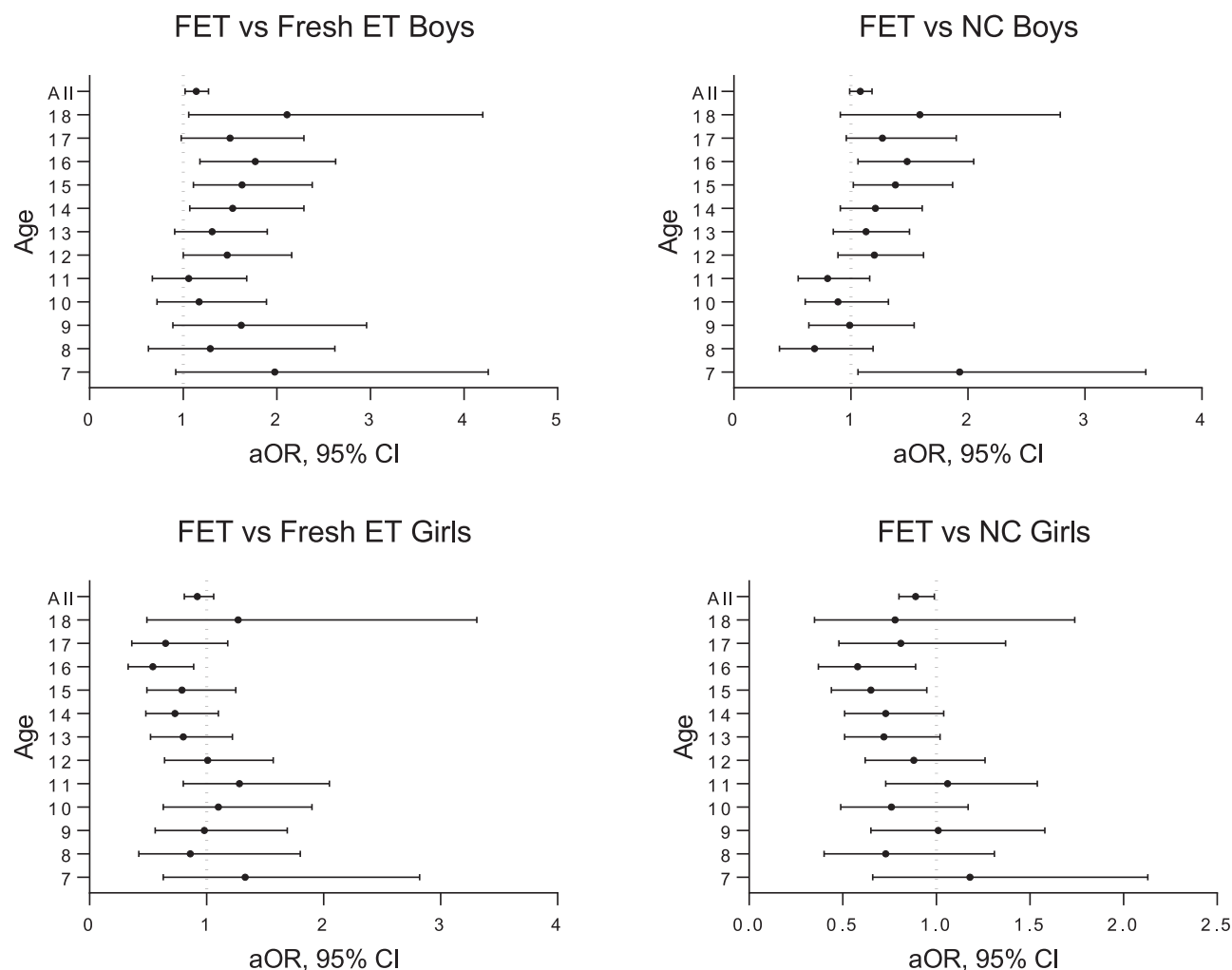
**Figure 2.** Forest plot of the adjusted odds ratios of overweight (ISO-BMI ≥ 25) for FET compared to fresh ET and FET compared to NC boys and girls by age. ISO-BMI, the age- and sex-adjusted BMI for children as described by the International Obesity Task Force (IOTF); FET, frozen embryo transfer; ET, embryo transfer; NC, natural conception; aOR, adjusted odds ratio; CI, confidence interval. Logistic regression. Adjusted for birth year, preterm birth, maternal age, parity, and socioeconomic status.

Most of the studies adjusted for various confounders, including height and/or weight of the parents (Ainsworth *et al.*, 2019; Turner *et al.*, 2020; Terho *et al.*, 2021b). However, parental anthropometric data were not available for adjustments in the study by Hann *et al.* (2018). The shorter follow-up time of these studies limits their comparability with our results.

A recent multi-cohort study using data from 26 population-based study cohorts (including singletons and multiples) analyzed the growth results in each cohort separately and subsequently combined the results as a meta-analysis. Fresh ET children tended to be shorter and lighter and FET children tended to be taller and heavier compared to NC children until the age of 6 to 9, although the differences were not statistically significant. Adjustments were made for maternal age, parity, BMI, smoking, education, ethnicity, offspring sex, and age. The number of FET children included in the analyses at different ages varied between 84 and 303 (Elhakeem *et al.*, 2022). A large Nordic register study on the risk of cardiovascular disease and diabetes found an increased risk of a diagnosis of childhood obesity for the ART compared to NC cohort (aHR 1.21, 95% CI 1.11–1.31; adjusted for sex, birth year, country, maternal age, and obesity). No significant differences between FET and fresh ET were found concerning obesity (Norrman *et al.*, 2021).

The longest follow-up on the growth of children after FET so far was published by a Norwegian research group. They compared growth of FET, fresh ET and NC children in two different register cohorts using self-reported data, the MoBa cohort reporting until age 7 and the Norwegian Army cohort at the age of 17. In their study, FET singletons (n = 239) were taller and heavier compared to fresh ET singleton for the first 2 years, but no significant differences were found in the cohort of 17-year-olds, including 176 FET children (Magnus *et al.*, 2021). The measurements at the age of 17 were adjusted for maternal age, parity, year of birth, sex, age at the time of self-reported measures of height and weight, and paternal height and BMI (Magnus *et al.*, 2021). Our results contradict the adolescent results presented by Magnus *et al.* (2021). It should be noted that in our study, the analyses were performed separately for boys and girls, whereas Magnus *et al.* (2021) analyzed the entire adolescent population, adjusting for sex.

Interestingly, in the present study, the findings were different for FET boys and girls. It has been suggested that the fetal risk of LGA associated with FET might be more strongly associated with male sex (Tang *et al.*, 2022). In our previous large cohort study based on birth weights by gestational week at birth, the risk of LGA birth weight was similar for FET boys and girls, although for
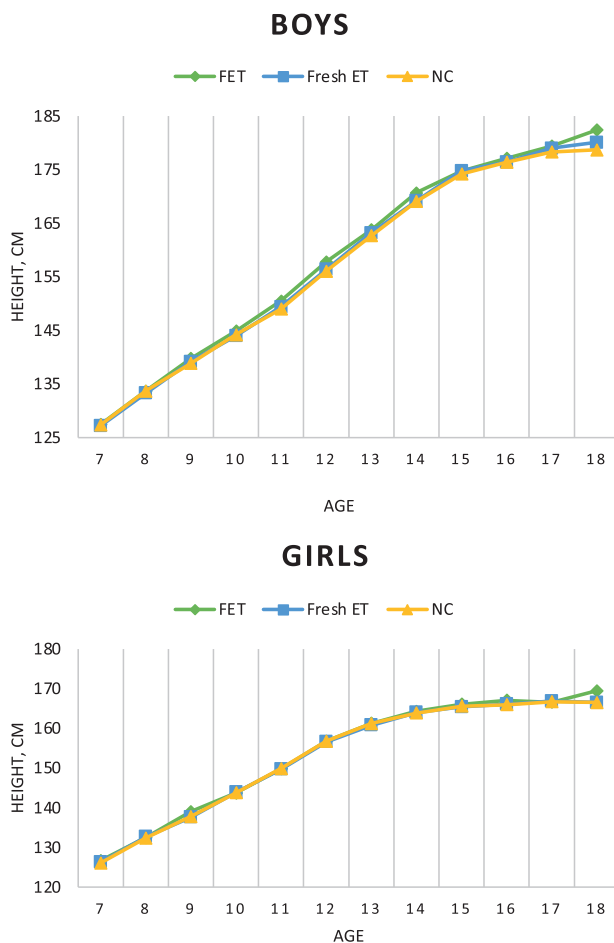
## BOYS



## GIRLS



**Figure 3. Adjusted heights of boys and girls by age.** FET, frozen embryo transfer; ET, embryo transfer; NC, natural conception. Adjusted for birth year, preterm birth, maternal age, parity, and socioeconomic status.

boys the mean birth weights were significantly higher compared to fresh ET boys from gestational week 33 and for girls the significant difference started from gestational week 34 (Terho *et al.*, 2021a). Other studies have reported the risk of LGA and high birth weight to associate more with FET boys compared to girls (Kaartinen *et al.*, 2015; Litzky *et al.*, 2018; Coetzee *et al.*, 2020; Tang *et al.*, 2022), suggesting male offspring may be more prone to the effects of the freeze-thaw process. In the results of the present study, highest proportion of LGA at birth was seen for boys. The results on growth suggest these changes might have an effect lasting until early adulthood. Unfortunately, within the size of this cohort, we were not able to stratify the growth measurements according to LGA at birth.

A possible mechanism to explain the different outcomes between FET, fresh ET, and NC are epigenetic changes, since ART procedures temporally overlap with significant epigenetic remodeling. A systematic review and meta-analysis suggest that ART is associated with aberrant DNA methylation in various tissues, but changes seen in placenta or cord blood do not necessarily persist long-term into childhood or adulthood (Barberet *et al.*, 2022). A large Norwegian cohort found differences in cord blood DNA methylation of genes related to growth, neurodevelopment, and disease for both FET (n = 126) and fresh ET (n = 764) compared to NC offspring. These changes were not explained by parental DNA methylation or subfertility and could be induced by

embryo freezing or the differences in the hormonal milieu of the uterus at the time of ET (Håberg *et al.*, 2022). Epigenetic changes may vary by age, tissue, or cell type. Interestingly, in the general population, also sex-specific DNA-methylome changes have been described in placenta, cord blood, and several other tissues (Bozack *et al.*, 2021; Andrews *et al.*, 2022; Grant *et al.*, 2022). However, the role these changes play in the different phenotypes of disease between males and females has not yet been adequately documented.

As puberty is an important determinant of growth velocity, differences in puberty timing might explain growth differences. In the present study, after adjustments, FET boys were taller than NC between 11 and 14 years of age, which might be explained by earlier onset of puberty in FET boys. However, a recent large Nordic study on puberty disorders of ART children suggests this is not likely. The risk of puberty disorders was increased for ART compared to NC children, but late onset of puberty was more common in ART boys, whereas early puberty was more common in ART girls compared to NC cohorts. Fresh ET offspring had a higher incidence of puberty disorders compared to FET offspring, whereas the adjusted risk did not significantly differ between fresh ET and FET compared to NC offspring (Klemetti *et al.*, 2022).

The Finnish growth charts, used in the primary health care throughout the country for screening of growth-related disorders, were renewed in 2010–2011 (Saari *et al.*, 2011). It should be noted that all mean weight and height measurements for the FET, fresh ET, and NC groups in the current study are well within the normal growth range between −2 and +2 SDs (data not shown). The clinical relevance of the small growth differences shown remains unknown. However, the increased risk of overweight in FET boys compared to fresh ET boys could be indicative of an increased risk of overweight or obesity in adulthood, as well as increased risks of cardiovascular disease, diabetes, musculoskeletal disorders, and some cancers (WHO, 2022). It should be noted, though, that BMI alone may not be a good indicator of cardiovascular risk, as it does not necessarily describe visceral adiposity.

The main limitation of this study is our incapability to adjust for parental anthropometric characteristics. Data on maternal prepregnancy weight and height are included in the FMBR since years 2004 (most hospitals)/2005 (all hospitals). Thus, we conducted a subanalysis on the maternal prepregnancy BMIs of the cohort children born between 2004 and 2006, including 698 FET and 837 fresh ET pregnancies. No significant differences in maternal BMIs were found between the study groups (data not shown). As for other limitations, unfortunately, growth data were not available for the whole cohort, but compared to previously published data on the growth of FET children, the numbers of FET children with available measurements at each age is reasonable in the present study. However, the numbers are limited at the start and end of the follow-up, thus these results should be regarded with caution. The size of our cohort allowed no analyses on the risk of severe obesity (ISO-BMI ≥ 35). Also, concerning obesity (ISO-BMI ≥ 30), the age-specific numbers of obese boys and girls were so small that they cannot be published due to regulatory constraints related to data protection and privacy. In addition, it should be considered that mainly cleavage stage embryos were transferred, and slow freezing was used in FET when the parents of this cohort were undergoing ART. Therefore, these results may not be generalizable to blastocyst transfers and vitrification. Data on the FET cycle modalities were unfortunately not available. We were able to adjust for several known

confounding factors; SES based on mothers' education among them. Since SES correlates strongly with maternal smoking, we did not include smoking as an independent factor in the adjustments (Rumrich *et al.*, 2018). Furthermore, we decided to adjust for PTB to better describe the possible effects of the FET and fresh ET treatments on child growth, independent of gestational age at birth. All analyses were also carried out without adjusting for PTB, but with adjustment for birth year, maternal age, parity, and SES. However, this did not change the significance of the results (data not shown).

A definite strength of this study are the reliable Finnish registers and our ability to link register data using the personal ID codes. Also, the growth measurements are collected prospectively by medical professionals, mainly nurses, working in the school healthcare system. We present the longest follow-up to date on the growth of FET children, and stratified the analyses based on the sex of the child instead of just adjusting for sex. Age- and sex-adjusted ISO-BMI for children was used to classify children with overweight. In addition, logistic regression was performed to investigate the odds of overweight between the groups.

## Conclusions

The risk of overweight among FET boys warrants further research as to verify the results in larger cohorts, and to further investigate the mechanisms behind this gender-specific finding. In addition, future ART research should aim to combine growth with long-term health data to unveil possible cardiometabolic risks associated with ART. The lack of significant differences in height after FET and fresh ET offers reassurance of the safety and feasibility of FET in the treatment of infertility.

## Supplementary data

Supplementary data are available at *Human Reproduction* online.

## Data availability

Under Finnish privacy laws, the study permission and access to register data were granted specifically to the authors. Upon reasonable request, statistical code and data output may be available from the corresponding author.

## Authors' roles

A.M.T., A.T., M.G., and S.P. planned the study. A.M.T. and M.G. carried out the statistical analyses. A.M.T. drafted the manuscript. All authors participated in the critical review of the analyses and the manuscript, and all authors approved the final version of the manuscript.

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## Conflict of interest

The authors declare no conflicts of interest.

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