Self-reported onset of puberty and subsequent semen quality and reproductive hormones in healthy young men

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Submitted on December 15, 2015; resubmitted on April 21, 2016; accepted on April 29, 2016

STUDY QUESTION: Is there an association between pubertal onset and subsequent reproductive health in young men?

SUMMARY ANSWER: Self-reported later onset of puberty was associated with reduced semen quality and altered serum levels of reproductive hormones among 1068 healthy, young Danish men.

WHAT IS KNOWN ALREADY: The long-term effects of variations in the onset of male puberty on subsequent reproduction remain largely unstudied.

STUDY DESIGN, SIZE, DURATION: In a cross-sectional study, young healthy Danish men were approached when they attended a compulsory medical examination to determine their fitness for military service from 2008 to 2012. A total of 1068 healthy, young Danish men (mean age 19 years) participated.

PARTICIPANTS/MATERIALS, SETTINGS, METHODS: They were asked to assess whether onset of penile and testicular growth, development of pubic hair and voice break occurred earlier, at the same time as or later than their peers. Their semen quality (semen volume, sperm concentration, total sperm count and percentages of motile and morphologically normal spermatozoa) and serum concentrations of sex hormones (LH, FSH, total testosterone, SHBG, inhibin B) and testicular size were determined.

MAIN RESULTS AND THE ROLE OF CHANCE: The response rate was 29%. Of the 1068 men who then participated, 652 answered the questions about penile growth and pubic hair development and were therefore included in the analysis. Self-reported later onset of puberty was associated with a 25% reduction in sperm concentration (95% CI −41%; −4%), a 40% reduction in total sperm count (−55%; −21%), a 1.6% age point reduction in morphologically normal spermatozoa (−2.9; −0.3) and a 1.6 ml reduction in testicular size (−2.4 and −0.8 ml), after adjustment for confounders. Self-reported later onset of puberty was also associated with a 9% (3%; 15%) reduction in free testosterone and a 16% (2%; 31%) increase in FSH, after adjustment for confounders.

LIMITATIONS, REASON FOR CAUTION: Our study was cross-sectional and reverse causality cannot be ruled out. In addition, we cannot rule out the possibility that the men with late puberty onset had not yet fully matured although most were in Tanner stage 5.

WIDER IMPLICATIONS OF THE FINDINGS: Approximately 15% of young Danish men have self-reported later onset of puberty than their peers. We found poorer testicular function in young men with a history of later pubertal development, suggesting that timing of pubertal onset may be a fundamental marker of male reproductive health. However, we cannot exclude the possibility that these men had not fully matured at the time of examination and therefore their semen quality may yet improve, which makes follow-up important.

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STUDY FUNDING/COMPETING INTERESTS: This work was supported by the Danish Council for Strategic Research, Program Commission on Health, Food and Welfare (project number 2101-08-0058), Rigshospitalet (grants 961506336 and R42-A1326), European Union, DEER (grant agreement no 212844), the Danish Ministry of Health and the Danish Environmental Protection Agency and Kirsten and Freddy Johansen Foundation (grant 95-103-72087). There are no competing interests.

Key words: puberty onset / male reproduction / semen quality / sex hormones / fertility

Introduction

Puberty represents the biological and psychological transition from childhood to adulthood. Several studies have reported that the age at pubertal onset has declined in girls (Sorensen et al., 2012), and newer studies (Ma et al., 2011; Herman-Giddens et al., 2012) suggest a similar trend for boys. A Danish study (Sorensen et al., 2010) found temporal trends in puberty onset in healthy boys from 1991–1993 to 2006–2008 with pubertal onset occurring 3 months earlier in 2006–2008 than in 1991–1993 (from 11.92 to 11.66 years). However, the results were not significant after adjustment for BMI.

A Danish study (Sorensen et al., 2010) found temporal trends in puberty onset in healthy boys from 1991–1993 to 2006–2008 with pubertal onset occurring 3 months earlier in 2006–2008 than in 1991–1993 (from 11.92 to 11.66 years). However, the results were not significant after adjustment for BMI.

To our knowledge, no previous studies have assessed the long-term influence of age at pubertal onset on subsequent male reproductive health in a normal population. It is a clinical experience that induction of spermatogenesis with gonadotrophin therapy takes much longer in men with a history of delayed puberty due to congenital hypogonadotropic hypogonadism (CHH) than in men who entered puberty at a normal time and pace but developed acquired pituitary insufficiency in adult life. This is likely to be due to differences in number of Sertoli cells, and in accordance with this, pretreatment with rFSH prior to hCG/GnRH has been successful in inducing testicular growth and fertility in men with CHH with prepubertal testes (Dwyer et al., 2013, 2015). Thus, we speculate that the timing of puberty and the associated testicular maturation may be important for fully mature reproductive capacity.

In a cross-sectional setting, we therefore examined the association between self-reported pubertal onset and subsequent reproductive health, measured as semen quality, testicular size and serum reproductive hormones, among 1068 normal, healthy Danish men.

Materials and Methods

Population

In Denmark, all men, except those with severe or chronic physical or psychological diseases (<15%), are required to attend a medical examination before being considered for military service. Men are called upon to present themselves at the age of 18–19 years, but some postpone this examination until completion of their education. The mean age at examination is 19 years and 87% are examined before age 21. Men attending the medical examinations are therefore considered representative of the general population of healthy young men. Since 1996 trained staff from the University Department of Growth and Reproduction (Rigshospitalet, Copenhagen, Denmark) have approached the young when they have appeared for the compulsory physical examination in the Copenhagen area (city area), independently of whether they were suited for military service or not, and invited them to participate in a study of semen quality. Men participating from January 2008 to June 2012 were included in the present study, as they completed a questionnaire including detailed information on puberty onset, which was not available before 2008. In addition to the questionnaire, all men delivered a semen sample, had a blood sample drawn and underwent a physical examination. They received financial compensation for their time (500 DKK to 70 Euro). Ethical approval was obtained from the local ethical committee. A detailed description of the study has previously been published (Jørgensen et al., 2012).

Semen analysis

All men provided a semen sample by masturbation in a room close to the semen laboratory. The period of ejaculation abstinence was recorded, and the semen sample was analysed for volume, sperm concentration, total sperm count, percentage of motile spermatozoa (A and B) and percentage of morphologically normal spermatozoa as described in Jørgensen et al. (2012), which is in accordance with the World Health Organization’s guidelines at the present time (World Health Organisation, 1999, 2010). The variation between technicians in measuring concentration was <10%. The same two experienced technicians assessed the sperm morphology according to strict criteria for all 904 microscopy slides (Menkveld et al., 1990).

Blood samples

Blood samples were drawn from a cubital vein, centrifuged and serum was separated and frozen. Serum levels of FSH, LH and sex hormone-binding globulin (SHBG) were determined using time-resolved immunofluorometric assays (Delfia, Wallac, Turku, Finland). Testosterone (T) levels were determined using a time-resolved fluorimunoassay (Delfia, Wallac, Turku). Intra- and inter-assay coefficients of variation (CVs) for measurements of FSH and LH were 3.0 and 4.5%, respectively. The CVs for T and SHBG were <8 and <5%, respectively. The intra- and inter-assay CVs for inhibin B were <7 and <6%, respectively. All hormones were measured within the same time period in the same assay batches. Free testosterone was calculated (cFT) based on the measured serum concentrations of total T and SHBG using the method of Vermeulen et al. (1999) and a fixed albumin concentration of 43.8 g/l. The T/LH ratio, the cFT/LH ratio and LH × T were calculated.

Physical examination

Physicians assessed the Tanner stage of pubic hair and genital development, testicular volumes, as determined by ultrasound (volume = L × W × H × 0.52 and mean of two testes calculated), the possible presence of a varicocele (grade 1–3) or hydrocele, the location of the testes (scoliotic or otherwise), and the consistency of the testis and epididymis. Conditions detected at the physical examination that could affect semen quality (such as varicocele grade 2 or 3 or abnormal position of the testes) were summarized in a
single variable: ‘Conditions found at the physical examination’. The weight and height of the men were also measured and body mass index (BMI) was calculated (weight in kilograms divided by squared height in metres).

**Questionnaire**

All participants completed a questionnaire prior to the physical examination including information on previous and/or current diseases including genital diseases such as inguinal hernia, varicocoele, epididymitis, gonorrhoea, chlamydia and surgery for testicular torsion. They were asked if they were born with both testicles in the scrotum. Self-reported diseases in the reproductive organs affecting semen quality were transformed into two variables: ‘Self-reported genital conditions’ (torsion of testes, epididymitis or inguinal hernia) and ‘Sexually transmitted diseases’ (STD, gonorrhoea or chlamydia). The participants were asked how frequently they had ejaculations (either related to intercourse, masturbation or involuntary), and responses were categorized into more or less often than every 5 days. In addition, they were asked how often they experienced a lack of interest in sex, difficulty in obtaining an erection, premature ejaculation, lack of ejaculation or other sexual problems; men were categorized as having sexually related problems if they had replied ‘often’ to one or more of these questions.

The men responded to a standard questionnaire about maternal education and exposure to smoking in utero. Their caffeine intake was assessed, as has previously been described (Jensen et al., 2010). A respondent was categorized as a smoker if he reported smoking more than once a week. The men were asked about daily alcohol consumption and their alcohol intake was calculated as the sum of daily reported unit intakes within the last week (categorized into below or above 21 units per week which was the higher limit for advised alcohol intake for males at the time from the Danish health authorities).

All men were asked when they experienced the onset of four distinctive pubertal changes: testicular growth, penile growth, pubic hair development and pubic hair. The response categories were: earlier than your peers, at the same time as your peers and later than your peers. The responses were studied separately and combined by creating three variables including men who replied early, average or late to respectively two, three or four pubertal changes: (i) penile growth and pubic hair development (available for n = 652); (ii) penile growth, pubic hair development and voice break (n = 487); (iii) penile growth, pubic hair development, voice break and testicular growth (n = 410). They were created in that order, as we believe that most men would recall penile growth, followed by pubic hair development, voice break and testicular growth.

**Statistics**

Outcome variables were semen volume, sperm concentration, total sperm count, percentage of motile and morphologically normal spermatooza, testosterone, free testosterone, FSH, LH, SHBG and inhibin B. Furthermore, testis size and T/LH ratio, cFT/LH ratio and LH x T were included. Exposure variables were the four distinctive pubertal changes: initiation of testicular or penile growth, initiation of voice change and development of pubic hair, as well as the combined answers divided into: early, average (reference) and late onset.

First, semen quality and reproductive hormones were compared for men in relation with their penile growth and development of pubic hair. Differences between the groups were tested by use of Kruskal Wallis test. We then compared the distributions of the variables from the questionnaires and physical examinations among men with different onsets of puberty by χ² test or one-way analysis of variance ANOVA for continuous variables in order to identify potential confounders (Table I). Finally, data were analysed using multiple linear regression analyses (Tables IV and V). Normally distributed outcome variables were entered directly as continuous variables in the model. Due to the non-normal (skewed) distributions of sperm concentration, total sperm count (values of zero were set to 0.1) and serum reproductive hormones, these variables were analysed on a natural logarithm scale and back-transformed to provide the percentage difference in the variables between men with respective early and late onset of puberty compared with average. Covariates initially included factors possibly associated with semen parameters, reproductive hormones or puberty onset and were then excluded stepwise if they did not change the estimate by more than 10%. The same set of confounders was used for the analyses of all semen parameters: period of abstinence (either below 48, 48–96, >96 h after which a plateau in association with sperm count was found), alcohol intake (≤21, >21 units per week), smoking, exposure to mother’s smoking in utero, STD and age (≤20, >20 years) and for sperm motility, time between ejaculation and analysis (less or more than 1 h). Tests size was not adjusted for period of abstinence. The analyses of reproductive hormones were adjusted for time of blood sampling, smoking and BMI (<20, 20–25 and ≥25 kg/m²).

As it is known that early onset of puberty is associated with adult BMI, BMI may be an intermediary factor between pubertal onset and reproductive health. We therefore performed the analyses both with and without adjustment for BMI. The analyses were also repeated among men in Tanner stage 5 (n = 553) only, as men with self-reported later onset of puberty may theoretically not be fully virilized at the time of examination.

The results are presented as regression coefficients with 95% confidence intervals. The fit of the models was evaluated by inspection of the residuals. All data were analysed in IBM SPSS statistics version 20.

**Results**

A total of 1243 men were examined, but 175 men were excluded from the present study for the following reasons: missing data on total sperm count and sperm concentration (n = 2), ejaculatory duct obstruction (n = 5), diagnosis of testicular cancer during the study (n = 1), previous treatment of malignant diseases (n = 2), previous surgery of vas deferens (n = 1), self-reported use of anabolic steroids (n = 10), no response to puberty questions (n = 154). Therefore, 1068 young men (mean age of 19.0 (range 18–28 years)) were included (for information, see Table I). Non-respondents were generally older than respondents but otherwise no differences in distribution of confounders were detected (data not shown). Five men had azosperma and were excluded from the semen analyses; however, only one of these five men had provided information on pubic hair development and voice break. A total of 25% and 20% reported early and late pubic hair development, and 17% and 16% reported voice break, 17% and 17% reported early and late penile growth and pubic hair development, whereas 13% and 12% reported early and late onset of penile growth, pubic hair development, voice break and testicular growth (Table II).

Men with earlier or later pubertal onset had poorer semen quality (Table III) and smaller testes. Early onset of puberty was associated with increased levels of free testosterone and lower levels of FSH and SHBG. Late onset of puberty was associated with higher levels of FSH, SHBG, and decreased levels of free testosterone (Table III) and lower LH x T levels.

The basic characteristics of the participating men can be seen from Table I. Men with self-reported early onset of puberty were shorter, had higher BMI, were more often smokers or exposed to smoking in utero and more often reported STDs and sexual problems than men who had average pubertal onset (Table I). Men with self-reported late onset of puberty were slimmer and taller, smoked less, were more often born with undecendent testis and more often reported sexual problems compared with men with average onset of puberty (Table I).

Self-reported early and late onset of penile growth and pubic hair development were associated with a reduction in semen quality after...
adjustment for confounders (Table IV). Sperm concentration was reduced by 16% (95% CI 2-34%; 7%) and 25% (2-42%; 25%) in men with early and late puberty, respectively, compared with average. In men with early and late puberty, total sperm count was reduced by respectively 17% (2-37%; 10%) and 40% (2-54%; 21%) and they had fewer morphologically normal spermatozoa by 0.4% point (2-1.6; 0.8) and 1.7% point (2-3.0; 0.4) and fewer motile spermatozoa by 1.9% point (2-5.6; 1.8) and 3.8% point (2-7.6; 20.1). Adjustment for BMI did not change the findings (Table IV). Men with early or late onset of puberty also had smaller testicles, respectively, by 2-0.6 ml (1.4 ml; 0.2 ml) and 2-1.7 ml (2-2.5 ml; 0.9 ml).

Late onset of penile growth and pubic hair development were associated with a decrease in free testosterone (9% (2-15%; 3%)) after adjustment, but due to an increase in SHBG the total testosterone did not differ. Men with late puberty also had lower LH levels (2-15% (2-28%; 15%), as well as a significant 16% increase in FSH (2%; 31%) and a tendency toward a lower LH × T (2-13 (2-25; 1)). No clear association between early pubertal onset and serum reproductive hormones was detected (Table V).

When studying the men who responded positively to each of the four puberty questions separately, similar associations were found, although the strongest association were for onset of testicular and penile growth (data not shown). In addition, the associations were strengthened when including only fully virilized (Tanner stage 5) men (data not shown).

**Discussion**

In this retrospective study of 19-year old healthy Danish men, we found that self-reported pubertal timing, especially delayed pubertal onset, was associated with a reduction in semen quality and testicular size, increased...
FSH levels, as well as decreased free testosterone and lower LH levels. This suggests that their testicular function may be affected and that the timing of pubertal onset may be a fundamental biomarker of overall male reproductive health. In addition, men with early onset of puberty more frequently had high-risk behaviour compared with men with average pubertal timing. Due to the cross-sectional design of our study, we cannot exclude reverse causality and cannot rule out the possibility that some of the men who reported late onset of puberty may not have been fully matured at the time of examination (as suggested by their higher levels of SHBG and lower BMI). There is therefore a possibility that their reproductive function will improve over time. However, restricting the analyses to fully mature men (Tanner stage 5) only strengthened our

### Table II  Respondents according to categories of pubertal onset.

<table>
<thead>
<tr>
<th>Puberty onset</th>
<th>All</th>
<th>Early</th>
<th>Average</th>
<th>Late</th>
</tr>
</thead>
<tbody>
<tr>
<td>Onset of penile growth</td>
<td>894</td>
<td>160</td>
<td>580</td>
<td>154</td>
</tr>
<tr>
<td>Onset of pubic hair development</td>
<td>1061</td>
<td>273</td>
<td>579</td>
<td>209</td>
</tr>
<tr>
<td>Onset of voice break</td>
<td>1068</td>
<td>183</td>
<td>713</td>
<td>67</td>
</tr>
<tr>
<td>Onset of testicular growth</td>
<td>737</td>
<td>117</td>
<td>527</td>
<td>72</td>
</tr>
<tr>
<td>Combined categories</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Penile growth and pubic hair development</td>
<td>652</td>
<td>114</td>
<td>430</td>
<td>108</td>
</tr>
<tr>
<td>Penile growth, pubic hair development and voice break</td>
<td>487</td>
<td>72</td>
<td>347</td>
<td>68</td>
</tr>
<tr>
<td>Penile growth, pubic hair development, voice break and testicular growth</td>
<td>410</td>
<td>53</td>
<td>308</td>
<td>49</td>
</tr>
</tbody>
</table>

### Table III  Semen quality and reproductive hormones among 652 young Danish men according to puberty onset defined by penile growth and development of pubic hair.

<table>
<thead>
<tr>
<th>Semen quality and reproductive hormones</th>
<th>Puberty onset</th>
<th>All</th>
<th>Early</th>
<th>Average</th>
<th>Late</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n = 652</td>
<td>n = 114</td>
<td>n = 430</td>
<td>n = 108</td>
<td></td>
</tr>
<tr>
<td></td>
<td>M 2.5–97.5</td>
<td>M 2.5–97.5</td>
<td>M 2.5–97.5</td>
<td>M 2.5–97.5</td>
<td></td>
</tr>
<tr>
<td><strong>Semen quality</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sperm concentration (mill/ml)</td>
<td>47</td>
<td>48</td>
<td>48</td>
<td>45</td>
<td></td>
</tr>
<tr>
<td>Total sperm count (mill)</td>
<td>145</td>
<td>133</td>
<td>156</td>
<td>131</td>
<td></td>
</tr>
<tr>
<td>Semen volume (ml)</td>
<td>3.2</td>
<td>3.2</td>
<td>3.2</td>
<td>3.1</td>
<td></td>
</tr>
<tr>
<td>Morphologically normal (%)</td>
<td>7.0</td>
<td>6.8</td>
<td>7.5</td>
<td>6.5</td>
<td></td>
</tr>
<tr>
<td>Motile sperm (%)</td>
<td>57</td>
<td>56</td>
<td>59</td>
<td>56</td>
<td></td>
</tr>
<tr>
<td>Testis size, ultrasonic (ml)</td>
<td>13.7</td>
<td>13.7</td>
<td>14.0</td>
<td>12.7</td>
<td></td>
</tr>
<tr>
<td>Period of abstinence (h)</td>
<td>62</td>
<td>63</td>
<td>62</td>
<td>62</td>
<td></td>
</tr>
<tr>
<td><strong>Reproductive hormones</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FSH (IU/l)</td>
<td>2.5</td>
<td>2.2</td>
<td>2.4</td>
<td>3.0</td>
<td></td>
</tr>
<tr>
<td>LH (IU/l)</td>
<td>3.4</td>
<td>3.5</td>
<td>3.4</td>
<td>3.2</td>
<td></td>
</tr>
<tr>
<td>Testosterone (nmol/l)</td>
<td>20.8</td>
<td>21.0</td>
<td>20.7</td>
<td>20.8</td>
<td></td>
</tr>
<tr>
<td>SHBG (nmol/l)</td>
<td>29</td>
<td>28</td>
<td>29</td>
<td>32</td>
<td></td>
</tr>
<tr>
<td>Free testosterone (pmol/l)</td>
<td>467</td>
<td>493</td>
<td>464</td>
<td>439</td>
<td></td>
</tr>
<tr>
<td>Inhibin B (pg/ml)</td>
<td>167</td>
<td>165</td>
<td>169</td>
<td>167</td>
<td></td>
</tr>
<tr>
<td>Estradiol (nmol/l)</td>
<td>81</td>
<td>82</td>
<td>80</td>
<td>77</td>
<td></td>
</tr>
<tr>
<td>Free T/LH ratio</td>
<td>149</td>
<td>152</td>
<td>149</td>
<td>136</td>
<td></td>
</tr>
<tr>
<td>Total T/LH ratio</td>
<td>6.3</td>
<td>6.2</td>
<td>6.3</td>
<td>6.3</td>
<td></td>
</tr>
<tr>
<td>LH × T</td>
<td>68.0</td>
<td>76.3</td>
<td>66.9</td>
<td>64.4</td>
<td></td>
</tr>
</tbody>
</table>

Median (M) and 2.5 and 97.5 percentiles.

*P < 0.05 Kruskal Wallis test.
### Table IV
Adjusted β coefficients from linear regression analyses indicating the absolute change in semen quality compared with reference group (for sperm concentration, total sperm count the percentage change is indicated) with 95% confidence intervals by onset of penile growth and pubic hair development.

<table>
<thead>
<tr>
<th>Puberty onset</th>
<th>n</th>
<th>Semen volume (ml)a</th>
<th>Sperm concentration %b</th>
<th>Total sperm count %b</th>
<th>Percent motile spermatozoa %b</th>
<th>Percent morphological normal spermatozoa %b</th>
<th>Testis size (ml)ab</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>β</td>
<td>95% CI</td>
<td>β</td>
<td>95% CI</td>
<td>β</td>
<td>95% CI</td>
<td>β</td>
</tr>
<tr>
<td>Early</td>
<td>114</td>
<td>0.0</td>
<td>−0.3; 0.3</td>
<td>−16</td>
<td>−34; 7</td>
<td>−17</td>
<td>−37; 10</td>
</tr>
<tr>
<td>Average</td>
<td>430</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
</tr>
<tr>
<td>Late</td>
<td>108</td>
<td>−0.3</td>
<td>−0.6; 0.1</td>
<td>−25</td>
<td>−42; −5</td>
<td>−40</td>
<td>−54; −21</td>
</tr>
<tr>
<td>Adjustment for BMI ab</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Early</td>
<td>114</td>
<td>0.0</td>
<td>−0.3; 0.3</td>
<td>−16</td>
<td>−34; 8</td>
<td>−16</td>
<td>−37; 10</td>
</tr>
<tr>
<td>Average</td>
<td>430</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
</tr>
<tr>
<td>Late</td>
<td>108</td>
<td>−0.3</td>
<td>−0.6; 0.1</td>
<td>−25</td>
<td>−41; −4</td>
<td>−40</td>
<td>−55; −21</td>
</tr>
</tbody>
</table>

Analyses are presented with and without adjustment for BMI.

aAdjusted for hours of abstinence (<48, 48–95 and >95 h), alcohol intake (>21 units per week), tobacco smoking, exposure to mother’s smoking in utero and age (>20 years).
bAdjusted also for duration between time of ejaculation and analysis of the sample.
cMeasured for 305 men.

dTransformed by the use of natural logarithm and back transformed giving the percentage change.

### Table V
Adjusted percentage change in reproductive hormones compared with reference group with 95% confidence intervals by onset of penile growth and pubic hair development.

<table>
<thead>
<tr>
<th>Puberty Onset</th>
<th>n</th>
<th>Testosterone (nmol/l)ab</th>
<th>Free testosterone (pmol/l)ab</th>
<th>FSH (IU/l)ab</th>
<th>SHBG (nmol/l)ab</th>
<th>Inhibin (pg/ml)ab</th>
<th>LH (IU/l)ab</th>
<th>FT/LH ratioab</th>
<th>T/LH ratioab</th>
<th>LH x testosteroneab</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>β</td>
<td>95% CI</td>
<td>β</td>
<td>95% CI</td>
<td>β</td>
<td>95% CI</td>
<td>β</td>
<td>95% CI</td>
<td>β</td>
<td>95% CI</td>
</tr>
<tr>
<td>Early</td>
<td>109</td>
<td>3</td>
<td>−4; 10</td>
<td>−7</td>
<td>−17; 5</td>
<td>−1</td>
<td>−9; 7</td>
<td>−3</td>
<td>−11; 5</td>
<td>6</td>
</tr>
<tr>
<td>Average</td>
<td>414</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
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<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
<td>Reference</td>
</tr>
<tr>
<td>Late</td>
<td>104</td>
<td>−5</td>
<td>−11; 2</td>
<td>−9</td>
<td>−15; −3</td>
<td>16</td>
<td>2; 31</td>
<td>8</td>
<td>0; 17</td>
<td>−4</td>
</tr>
</tbody>
</table>

aAdjusted for BMI levels (<20, 20–25 and >25), tobacco smoking, hour of day of blood sampling.
bTransformed by the use of natural logarithm and back transformed giving the percentage change.
findings. Furthermore, a follow-up study of 158 young Danish men recruited in 2005 showed that semen quality was unchanged during a 4-year follow-up, indicating that the men had fully mature capacity for sperm production (Carlsen et al., 2005).

In a newly published study, self-reported puberty was reported among more than 450,000 individuals. Early or late onset of menarche or voice break was associated with increased risks of a wide range of diseases including diabetes and cardiovascular disease (Day et al., 2015) and previous studies have suggested association of timing of voice break to later puberty onset on reproductive health among healthy men. However, our knowledge, no studies have assessed the long-term effects of obesity (Ong et al., 2012) and blood pressure (Hardy et al., 2006). To our knowledge, no studies have assessed the long-term effects of puberty onset on reproductive health among healthy men. However, precocious puberty has known long-term mental and physical complications in females (Sippell, 1994) such as a higher prevalence of risk behaviour introducing them earlier to alcohol, smoking and sexual debut and one study (Bratberg et al., 2007) found associations between perceived pubertal timing and increased risk behaviour for both sexes, although that study did not include any physical or biochemical assessments.

A few studies have reported that men with delayed puberty onset have decreased bone mineral density, increasing their risk of osteoporosis (Finkelstein et al., 1992; Bertelloni et al., 1995; Luboshitzky et al., 1998; Waugh et al., 2009). Additionally height and growth velocity have been found to be compromised in children with delayed puberty onset (Poyrazoglu et al., 2005). Several studies (Christiansen and Skakkebaek, 2002; Pitteloud et al., 2002; Raivio et al., 2007) of men with hypogonadotrophic hypogonadism with delayed puberty have shown that despite treatment with gonadotrophins or recombinant FSH sufficient to induce puberty and spermatogenesis, these men have sub-optimal semen quality. A study (Lemcke et al., 1996) of long-term effects of adolescent testosterone-one treatment for excessively tall stature reported a significant reduction in sperm motility a non-significantly lower sperm concentrations and total sperm counts as well as reduced normal sperm morphology. Although the authors suggest that these differences may be attributed to an excess of varicocele and maldescended testicles among the testosterone-treated men, their findings could also be explained by impaired testicular development. This suggests that the timing of the onset of pubertal development may be important for subsequent reproductive function.

Our study has potential public health interest because it was conducted among young, healthy men from the general population. Our participation rate was ~30%, which is higher than in similar cross-sectional studies (Swan et al., 2003; Iwamoto et al., 2013). Almost all of the men (93%) were unaware of their own fertility potential, making this unlikely to have affected their motivation to participate. In a previous study, we compared hormone levels among the participants and non-participants and found no significant difference with regard to reproductive hormones levels in the two groups, indicating that our participants represented the general population of young Danish men with respect to reproductive health (Andersen et al., 2000; Jorgensen et al., 2012). However, up to 15% of young men, suffering severe or chronic physical or psychological diseases, do not attend the examination and therefore our study population represents the healthy population of young men. These exclusions are not necessarily associated with pubertal development.

Our study was cross-sectional and we relied on self-reported, retrospectively assessed onset of puberty. Therefore, instead of asking the men to specify a certain age of pubertal onset, we asked them to place themselves in comparison with same-aged peers, as they are more likely to recall whether they entered puberty at the same time as their peers or earlier or later. Prospectively assessed self-reported timing of voice break has been found to be a fairly accurate non-invasive measure of pubertal maturation (Ong et al., 2012) but self-assessment of Tanner stage has been found to be inferior to clinical assessment (Faria et al., 2013). However, to our knowledge Tanner stage has not been validated when reported by recall in adult life. In a study among approximately 200,000 men aged 40–69 years, 4.3 and 5.9%, respectively, reported that they were younger or older than their peers at voice break (Day et al., 2015) which is considerably fewer than our 17% and 16%. However, our men were between 18 and 28 years and therefore had a shorter length of recall and may therefore more accurately recall their age at voice break. However, some young men may feel they are not virile enough which may have coloured their subjective recollections of pubertal milestones. To our knowledge self-reported, retrospectively assessed timing of testicular growth, penile growth and development of pubic hair has not been validated. Interestingly, the percentage reporting early or late onset of voice break, penile and testicular growth were approximately similar indicating the same magnitude of recall problems whereas more reported early or late onset of pubic hair development indicating that this may be more inaccurately assessed. We assumed that it is generally easier to recall changes that are visual, such as penile growth and development of pubic hair, and this assumption formed the basis of our analysis with categorization of men according to the number of pubertal questions answered consistently. As the men were unaware of their semen quality and reproductive hormones when responding to the questions, it is unlikely to have affected their response thereby leading to a non-differential misclassification underestimating the true effect. In addition, the findings were consistent for all four signs of pubertal onset assessed individually as well as the categorized responses.

Because high-risk behaviour has been associated with early puberty onset, primarily observed in girls (Bratberg et al., 2007), we suspected that our findings in the early onset group could be explained by their smoking, and sexual habits. We adjusted for these known risk factors, which did not change the finding. However, residual confounding from other behavioural or lifestyle factors cannot be ruled out. Since BMI has been associated with early puberty onset in girls (Wang, 2002; Rosenfield et al., 2009), and although data have not been conclusive with regard to boys (Ribeiro et al., 2006; Lee et al., 2010; Sorensen et al., 2010), BMI may be an intermediary factor. We did the analyses both with and without BMI adjustment, with essentially the same findings. In addition, BMI was lower among men with late onset of puberty, suggesting that they may not have fully developed their muscular strength.

In conclusion, in this cross-sectional study, we found poorer testicular function (semen quality, Leydig and Sertoli cell function) in young men with a history of self-reported later pubertal development, suggesting that timing of pubertal onset may be a fundamental marker of male reproductive health. We cannot rule out the possibility that the men with late onset of puberty were not fully mature at the time of examination despite their age. Therefore, there is a possibility that their reproductive function will improve over time. Our study is the first to report associations between pubertal timing and adult reproductive function and does not support that treatment of early or late puberty will have beneficial effects, but rather suggests that more work is needed to elucidate the mechanisms of pubertal timing to later reproductive health.
Authors’ roles

T.K.J. and K.F.F. performed data analysis and interpretation and drafted the manuscript. A.J., N.J., and N.E.S. provided assistance with data analysis and revised and edited the manuscript. L.A.O., L.N., L.P., U.N.J., A.K.B., M.K. and A.M.A. performed data collection, provided assistance with data analysis and interpretation, and revised and edited the manuscript. All authors revised and edited the manuscript.

Funding

This work was supported by the Danish Council for Strategic Research, Program Commission on Health, Food and Welfare (project number 2101-08-0058), Rigshospitalet (grants 961506336 and R42-A1326), European Union, DEER (grant agreement no 212844), the Danish Ministry of Health and the Danish Environmental Protection Agency and Kirsten and Freddy Johansens Foundation (grant 95-103-72087). Innovation Fund Denmark (14-2013-4).

Conflict of interest

None declared.

References


