

RESEARCH REVIEW

Minipuberty in Klinefelter syndrome: Current status and future directions

Lise Aksglaede¹ | Shanlee M. Davis^{2,3} | Judith L. Ross^{4,5} | Anders Juul¹

¹Department of Growth and Reproduction, Rigshospitalet, University of Copenhagen, Copenhagen, Denmark

²Department of Pediatrics, University of Colorado School of Medicine, Aurora, Colorado

³eXtraordinarY Kids Program, Children's Hospital Colorado, Aurora, Colorado

⁴Department of Pediatrics, Thomas Jefferson University, Philadelphia, Pennsylvania

⁵DuPont Hospital for Children, Wilmington, Delaware

Correspondence

Lise Aksglaede, Department of Growth and Reproduction, GR, 5064, Rigshospitalet, Blegdamsvej 9, DK-2100 Copenhagen Ø, Denmark.
Email: lise.aksglaede@regionh.dk

Funding information

Foundation for the National Institutes of Health, Grant/Award Number: K23HD092588

Abstract

Klinefelter syndrome is highly underdiagnosed and diagnosis is often delayed. With the introduction of non-invasive prenatal screening, the diagnostic pattern will require an updated description of the clinical and biochemical presentation of infants with Klinefelter syndrome. In the first months of life, the hypothalamic–pituitary–gonadal (HPG)-axis is transiently activated in healthy males during the so-called minipuberty. This period represents a “window of opportunity” for evaluation of the HPG-axis before puberty and without stimulation tests. Infants with Klinefelter syndrome present with a hormonal surge during the minipuberty. However, only a limited number of studies exist, and the results are contradictory. Further studies are needed to clarify whether infants with Klinefelter syndrome present with impaired testosterone production during the minipuberty. The aim of this review is to describe the clinical and biochemical characteristics of the neonate and infant with Klinefelter syndrome with special focus on the minipuberty and to update the clinical recommendations for Klinefelter syndrome during infancy.

KEYWORDS

Klinefelter syndrome, minipuberty, testosterone, XXY

1 | INTRODUCTION

Patients with Klinefelter syndrome (47,XXY) are diagnosed throughout life. In our single-center evaluation of approximately 200 patients with Klinefelter syndrome in Denmark, 20% were diagnosed prenatally, 30% were diagnosed during childhood due to excessive growth and/or behavioral problems, and the remaining half were diagnosed in adulthood, mainly as part of infertility evaluation (Aksglaede et al., 2011). Importantly, Klinefelter syndrome is highly underdiagnosed, and because of subtle symptoms before puberty diagnosis is often delayed (Bojesen et al., 2003; Berglund et al., 2019). Until recently, Klinefelter syndrome was rarely detected prenatally and always as an incidental finding during invasive diagnostic testing by chorionic villus sampling or amniocentesis because of increased risk for autosomal aneuploidy or advanced maternal age.

With the introduction of non-invasive prenatal screening (NIPS) it has become possible to detect fetal aneuploidies including sex

chromosome aneuploidies (SCA), based on genetic analysis of cell-free DNA isolated from maternal blood (Bianchi & Wilkins-Haug, 2014). Despite limitations (e.g., false positive results) in the detection of SCAs by NIPS (Gil et al., 2017), some countries offer routine NIPS including screening for SCA. Klinefelter syndrome may therefore be identified prenatally more frequently because of suspected SCA on NIPS.

A recent Australian study of the trends in prenatal diagnosis of SCAs before and after the introduction of NIPS showed that SCAs now represent a larger proportion of the prenatal results. However, the prenatal prevalence of SCAs as a percentage of births remains unchanged due to a decline in invasive prenatal diagnostic testing (Howard-Bath et al., 2018). These positive fetal screening results were not followed antenatally, therefore it is unknown how many were confirmed positive after birth. More advanced techniques may also improve the sensitivity and specificity in the detection of SCAs by NIPS, and the future prenatal diagnostic pattern of Klinefelter

syndrome may therefore necessitate novel updated clinical management plans for the care of these infants.

The aim of this review is therefore to describe the clinical and biochemical characteristics of the neonate and infant with Klinefelter syndrome with special focus on the minipuberty, and to update the clinical recommendations for Klinefelter syndrome during infancy.

2 | MINIPUBERTY IN HEALTHY MALE INFANTS—A “WINDOW OF OPPORTUNITY”

In healthy males the hypothalamic–pituitary–gonadal (HPG)-axis is transiently activated in the second trimester of fetal life (Clements et al., 1976; Kaplan & Grumbach, 1976; Massa et al., 1992; Winter, 1982) and in the first months of life during the so-called minipuberty (Andersson et al., 1998; Forest et al., 1974; Winter et al., 1975, 1976). Minipuberty is followed by a relatively quiescent period until reactivation at puberty when secondary sexual characteristics develop, and reproductive capacity is attained. The biological purpose for this transient activation of the entire HPG-hormone-axis in the first months of life remain unknown. It is believed to be fundamental for the normal development of male reproductive organs including completion of testicular descent, and may potentially contribute to sexual differentiation of multiple tissues, including modulating masculine neurobehavioral development (Alexander, 2014; Boas et al., 2006; de Mello et al., 2012; Dkhil et al., 2015; Ghahramani et al., 2014; Hines et al., 2016; Kuiri-Hanninen et al., 2011; Kuiri-Hanninen et al., 2014; Kuiri-Hanninen et al., 2019; Kung et al., 2016; Lamminmaki et al., 2012; Lanciotti et al., 2018; Main et al., 2000; Nugent et al., 2015; Pasterski et al., 2015; Swift-Gallant et al., 2016). One study demonstrated that testosterone during the first months of life correlated with an increase in longitudinal growth, accounting for ~15% of sex differences in adult height (Kiviranta et al., 2016). Furthermore, boys with congenital hypogonadotropic hypogonadism (CHH) appear to have diminished linear growth during minipuberty (Varimo et al., 2016). The mini-puberty period also appears to be a key time that lifelong sex differences in body composition begin, with males accumulating more lean mass and ultimately lower percent fat mass than females (Davis et al., 2019). There is much to learn about the immediate and programming effects of this transient HPG activation (Copeland & Chernauek, 2016).

In healthy male infants the concentrations of luteinizing hormone (LH) and follicle stimulating hormone (FSH) are low in cord-blood, but both increase shortly after birth, as the suppressive effects of placental hormones (especially estrogens) disappear (Schmidt & Schwarz, 2000). The resulting activation of the testicles leads to increasing secretion of testosterone (T), inhibin B, anti-Müllerian hormone (AMH), and insulin-like3 (INSL3) (Aksglaede et al., 2010; Andersson et al., 1998; Bay et al., 2007; Bergada et al., 2006; Forest et al., 1980). LH peaks at 2–10 weeks of age and decreases to prepubertal concentrations at 4–6 months of age, whereas T concentrations peak at 1–3 months and decline by 6 months of age. In line with the concentration of FSH, AMH, inhibin B and INSL3 also increase. AMH

peaks during the minipuberty, followed by a slight decline around 12 months, but remains elevated until puberty, whereas inhibin B decreases slightly around 15 months of age (Aksglaede et al., 2010; Bergada et al., 2006; Kuiri-Hanninen et al., 2014; Kuiri-Hanninen et al., 2019; Lanciotti et al., 2018). After the minipuberty period, LH, FSH, and T all decline to very low concentrations until reactivation of the HPG-axis at the time of central puberty. Inhibin B and AMH decrease less dramatically and remain at measurable concentrations during the quiescent childhood period, seemingly independent of gonadotropin concentrations.

During the transient activation of the HPG-axis in infancy, the number of Leydig cells increases, but after 3 months of age, the number gradually declines due to apoptosis of fetal Leydig cells (Codesal et al., 1990). Likewise, the Sertoli cells start to develop and the number of germ cells increases (Cortes et al., 1987; Muller & Skakkebaek, 1984), but because of lack of androgen receptors, spermatogenesis is not initiated (Boukari et al., 2009). Also related to the absence of androgen receptors at this time point, there is no inhibitory effect on the Sertoli cells, and the secretion of AMH remains high, despite high T concentrations. Clinically, testicles and penis increase in size during this period, although the testicular volume decreases slightly after the minipuberty (Boas et al., 2006; Kuiri-Hanninen et al., 2011; Main et al., 2006).

During the quiescent period following minipuberty the reproductive hormones are suppressed to very low or unmeasurable concentrations by most conventional immunoassays. The minipuberty, therefore, represents a “window of opportunity” for evaluation of the HPG-axis before puberty and without stimulation tests (for review, Grumbach, 2005). Knowledge about the reproductive hormones during the minipuberty is interesting from a physiological point of view, but it is also relevant from a diagnostic point of view. However, only limited studies from this period exist. Evaluation of the reproductive hormones during male minipuberty is relevant in infants with suspected CHH or differences in sex development (DSD) (Bouvattier et al., 2002; Johannsen et al., 2018; Kaiserman et al., 1998). If the diagnosis of primary versus secondary hypogonadism is not identified in infancy, the diagnosis may be delayed until adolescence or even adulthood.

3 | TREATMENT OF HYPOGONADISM IN MINIPUBERTY

In addition to an opportunity to evaluate the HPG-axis, the minipuberty period has been proposed as a time-sensitive opportunity for testosterone treatment in conditions with an impaired HPG-axis. Most experience about testosterone treatment during infancy comes from the treatment of micropenis, which can occur with either primary or secondary hypogonadism. Traditionally this treatment includes a short course of testosterone supplementation during minipuberty, an effective and well-tolerated regimen (Bin-Abbas et al., 1999; Hatipoglu & Kurtoglu, 2013) without virilization or disturbances of growth. Testosterone enanthate or cypionate is typically

used at the dose of 25–50 mg/month for 3 months. Treatment may either be administrated as intramuscular injections, topical application or suppositories.

Treatment with testosterone is effective with regards to penile growth if the tissue has normal androgen sensitivity. A few reports of infants with hypogonadotropic hypogonadism treated with gonadotropin during minipuberty exist (Bougnères et al., 2008; Main et al., 2002; Stoupa et al., 2017). These indicate that short-term treatment with recombinant FSH and LH may mimic the physiological minipuberty by stimulating the production of T, inhibin B and AMH, and potentially stimulating the proliferation of Sertoli cells (for review Swee & Quinton, 2019). However, treatment with gonadotropin agonists would not likely be beneficial in primary hypogonadism.

4 | MINIPUBERTY IN KLINEFELTER SYNDROME

Only a limited number of studies of the minipuberty in Klinefelter syndrome exists (Aksglaede et al., 2007; Cabrol et al., 2011; Lahlou et al., 2004; Ross et al., 2005), and the first publications from 2004 to 2007 were limited by low numbers of boys (total of 30 infants [$n = 10$, $n = 10$, and $n = 10$]; Aksglaede et al., 2007; Lahlou et al., 2004; Ross et al., 2005). The results of the first studies using sensitive immunoassays were contradictory, with one study reporting high-normal T (Aksglaede et al., 2007), and the others reporting lower than expected T (Lahlou et al., 2004; Ross et al., 2005).

A more recent study including 38 infants with Klinefelter syndrome presented detailed biochemical evaluation of the HPG-axis including T measured by means of liquid chromatography/tandem mass spectrometry (LC-MS/MS), which is now considered gold standard (Cabrol et al., 2011; Lahlou et al., 2011). In that study, T concentrations turned out to be significantly lower in the infants with Klinefelter as compared with controls, although the majority of the infants with Klinefelter presented with T concentrations within the low normal range. In contrast, INSL3 and LH concentrations were normal, and LH correlated positively with INSL3 and T. T and INSL3 are both markers of the Leydig cell function, and the result indicates that the Leydig cells are sensitive to LH action, although some degree of Leydig cell insufficiency during minipuberty, at least based on the findings about T, cannot be excluded (Cabrol et al., 2011; Lahlou et al., 2011).

In the study by Cabrol et al., (2011) FSH concentrations were significantly elevated in the infants with Klinefelter despite normal inhibin B. In a subset of these infants (17%) inhibin B was reduced despite normal FSH, and in 25%, inhibin B was normal despite elevated FSH. The authors reported a positive correlation between inhibin B and FSH, and that the Sertoli cells may be at least relatively resistant to FSH in some of these infants. AMH concentrations were in the normal range during minipuberty. There was no correlation between AMH and FSH or T, which is in accordance with the absence of androgen receptors on the Sertoli cells.

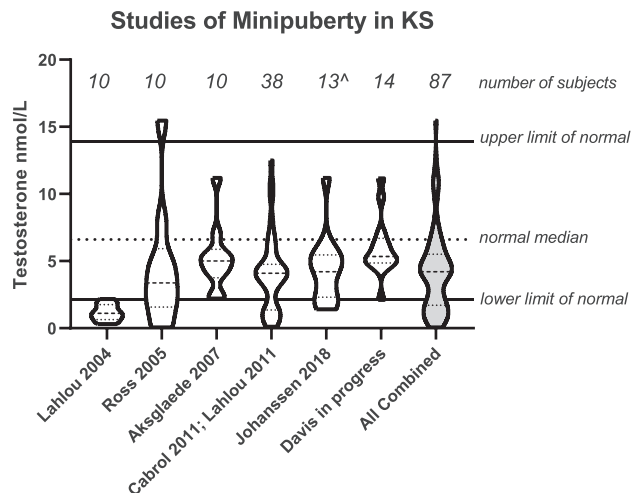


FIGURE 1 Summary of results from studies reporting testosterone concentrations in the minipuberty period in infants with Klinefelter syndrome. Violin plots display the median, interquartile range, and the distribution with the entire range for each study and with all the data combined (gray). Normal values are estimated from laboratory reference ranges and publications. Nine of these subjects were published in Aksglaede et al., 2007 and were not duplicated in the combined summary

Accepting the limitations inherent in combining studies with different methods, Figure 1 demonstrates previously reported testosterone concentrations during the minipuberty in infants with KS. Taken together, the total number of boys with Klinefelter syndrome represented in these studies on serum hormone concentrations during the first 6 months of life is <100. All the studies conclude that activation of the HPG-axis does occur, but whether or not differences in testicular hormone production exist needs further investigation.

5 | CLINICAL PRESENTATION IN INFANTS WITH KLINEFELTER SYNDROME

Features of Klinefelter syndrome in the prepubertal period are variable and there are no symptoms that are present in all diagnosed cases. Birth length, weight and head-circumference are generally within normal limits although slightly smaller than in males without Klinefelter syndrome (Aksglaede et al., 2008; Cabrol et al., 2011; Lahlou et al., 2011; Ross et al., 2005). Subtle dysmorphic signs such as fifth finger clinodactyly, high-arched palate, epicanthal folds and hypertelorism may be observed (Ross et al., 2005; Zeger et al., 2008) but none of these symptoms are pathognomonic. Congenital anomalies such as renal malformations, cardiac defects, cleft lip or palate, and club foot are rarely reported in infants with Klinefelter syndrome. Infants with Klinefelter syndrome are often noted to have some degree of hypotonia as well as an overall passive temperament. Developmental delays are common and most receive early intervention services (Robinson et al., 1990; Tartaglia et al., 2020).

Micropenis and cryptorchidism are found more frequently in infants with Klinefelter syndrome as compared with boys with a normal karyotype (Bojesen et al., 2006), however they are still uncommon (<10%) (Ross et al., 2005). Reduced penile length and testicular volume have been reported in most of the existing studies of infants with Klinefelter syndrome, and growth of the penis between infancy and childhood has also been reported as sub-normal (Davis et al., 2019; Lee et al., 2007; Ratcliffe et al., 1986; Ross et al., 2005; Zeger et al., 2008; Zinn et al., 2005).

Only a few studies of testicular histology from fetuses and neonates with Klinefelter syndrome exist. These have shown that the number of germ cells is reduced already at this early age, whereas the number of Leydig cells appear normal (Autio-Harmainen et al., 1980; Coerdts et al., 1985; Murken et al., 1974; Ratcliffe, 1982).

6 | TESTOSTERONE TREATMENT DURING MINIPUBERTY IN KLINEFELTER SYNDROME

Based on the findings of T concentrations in the lower range of normal during minipuberty and the reduced penile length and testicular volume suggesting testicular dysfunction, T replacement has been offered to an increasing number of infants with Klinefelter syndrome. Samango-Sprouse et al. retrospectively evaluated the neurodevelopmental effects of T replacement (monthly injection of 25 mg testosterone enanthate for 3 months) in infants with Klinefelter syndrome aged 4–15 months. At age 36 and 72 months the treated group was compared to an untreated group, and the treated group had higher scores in some domains of behavior, anxiety, neuromotor, speech and language, intellectual, and reading function (Samango-Sprouse et al., 2013, 2015, 2019). Importantly, some limitations of these studies should be noted. They were retrospective, nonrandomized, and likely subject to ascertainment bias, and the boys were treated at different ages with the majority being treated after the minipuberty.

One randomized study of T replacement in Klinefelter syndrome during the minipuberty, including 20 infants who were randomized to either no treatment or treatment with testosterone cypionate 200 mg/ml IM injection of 25 mg (0.125 ml) every 4 weeks for a total of three doses was recently published. Excess accumulation of adiposity was observed in the untreated boys, whereas the treated boys had normal body composition measures at 5 months of age. In the treated group linear growth and stretched penile length increased significantly, whereas no difference in testicular volume was observed (Davis et al., 2019). A larger, randomized and placebo-controlled study is underway to investigate the potential beneficial or unanticipated side effects of testosterone in infants with Klinefelter syndrome (PI Davis, NCT03325647).

7 | RECOMMENDATIONS

Until more data emerge, it is difficult to provide evidence-based recommendations to guide clinical care in infants with Klinefelter

syndrome. However, based on our experience and the limited studies summarized in this review, we provide several considerations for clinicians:

- Infants identified prenatally should have a postnatal chromosome analysis either on cord blood or peripheral blood to confirm prenatal findings, particularly if only a prenatal screening test (i.e., cell-free fetal DNA) was obtained.
- Clinical examination, assessing for any dysmorphic features, congenital anomalies, hypotonia, cryptorchidism, and micropenis should be conducted by an experienced medical provider.
- Given ~1/4 of infants with Klinefelter syndrome will have a testosterone concentration below the normal range (Figure 2), measurement of serum testosterone by LC-MS/MS at ~2 months of age (between 1 and 5 months) can be considered for additional information of individual testicular function. Measuring testosterone in infants older than 6 months of age is not useful.
- If micropenis (<2.5 standard deviations below the mean for age) is present, a short course of testosterone can be considered to improve functional limitations.
- Testosterone treatment in the absence of micropenis should not be considered standard of care given the limited evidence, but may be considered on an individual basis. If pursued, this should be done under the guidance of a medical provider with training and experience in prepubertal testosterone treatment, and the infant should be appropriately monitored for any side effects.

The ideal study of the minipuberty in Klinefelter syndrome would include an adequately powered cohort study of infants with Klinefelter syndrome and age-matched healthy controls, comparing biochemical measurements of LH, FSH, inhibin B, INSL3, AMH, and T

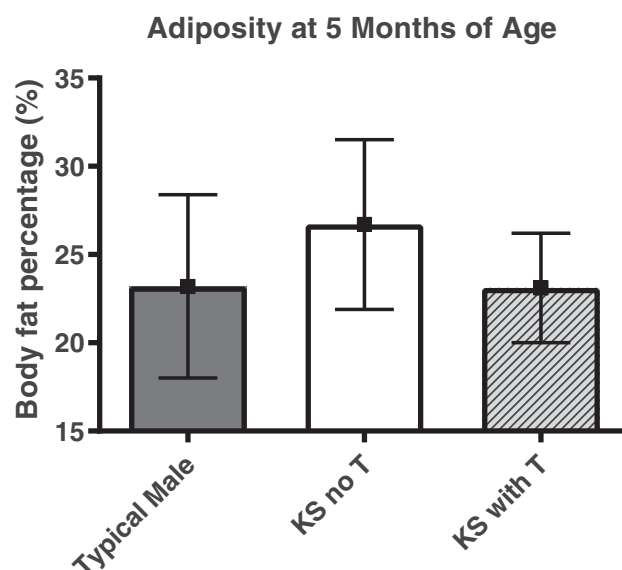


FIGURE 2 Testosterone treatment normalizes body composition in infants with Klinefelter syndrome. Data are mean and standard deviations

(measured by LC–MS/MS), all measured in the same lab. Ideally these infants would also be followed over time to assess whether there is a relationship between early hormonal concentrations and the physical and neuropsychological manifestations of Klinefelter syndrome.

ACKNOWLEDGMENTS

Shanlee Davis was supported by K23HD092588. Contents are the authors' sole responsibility and do not necessarily represent official NIH views.

ORCID

Lise Aksglaede  <https://orcid.org/0000-0001-7885-7467>

Shanlee M. Davis  <https://orcid.org/0000-0002-0304-9550>

Anders Juul  <https://orcid.org/0000-0002-0534-4350>

REFERENCES

- Aksglaede, L., Skakkebaek, N. E., Almstrup, K., & Juul, A. (2011). Clinical and biological parameters in 166 boys, adolescents and adults with nonmosaic Klinefelter syndrome: A Copenhagen experience. *Acta Paediatrica*, 100(6), 793–806. <https://doi.org/10.1111/j.1651-2227.2011.02246.x>
- Aksglaede, L., Sorensen, K., Boas, M., Mouritsen, A., Hagen, C. P., Jensen, R. B., ... Juul, A. (2010). Changes in anti-Mullerian hormone (AMH) throughout the life span: A population-based study of 1027 healthy males from birth (cord blood) to the age of 69 years. *Journal of Clinical Endocrinology and Metabolism*, 95(12), 5357–5364. <https://doi.org/10.1210/jc.2010-1207>
- Aksglaede, L., Petersen, J. H., Main, K. M., Skakkebaek, N. E., & Juul, A. (2007). High normal testosterone levels in infants with non-mosaic Klinefelter's syndrome. *European Journal of Endocrinology*, 157(3), 345–350. <https://doi.org/10.1530/EJE-07-0310>
- Aksglaede, L., Skakkebaek, N. E., & Juul, A. (2008). Abnormal sex chromosome constitution and longitudinal growth: Serum levels of insulin-like growth factor (IGF)-I, IGF binding Protein-3, luteinizing hormone, and testosterone in 109 males with 47,XXY, 47,XYY, or sex-determining region of the Y chromosome (SRY)-positive 46,XX karyotypes. *Journal of Clinical Endocrinology and Metabolism*, 93(1), 169–176. <https://doi.org/10.1210/jc.2007-1426>
- Alexander, G. M. (2014). Postnatal testosterone concentrations and male social development. *Frontiers in Endocrinology (Lausanne)*, 5, 15. <https://doi.org/10.3389/fendo.2014.00015>
- Andersson, A. M., Toppari, J., Haavisto, A. M., Petersen, J. H., Simell, T., Simell, O., & Skakkebaek, N. E. (1998). Longitudinal reproductive hormone profiles in infants: Peak of inhibin B levels in infant boys exceeds levels in adult men. *Journal of Clinical Endocrinology and Metabolism*, 83(2), 675–681. <https://doi.org/10.1210/jcem.83.2.4603>
- Autio-Harmainen, H., Rapola, J., & Aula, P. (1980). Fetal gonadal histology in XXXXY, XYY and XXX syndromes. *Clinical Genetics*, 18(1), 1–5. <https://doi.org/10.1111/j.1399-0004.1980.tb01356.x>
- Bay, K., Virtanen, H. E., Hartung, S., Ivell, R., Main, K. M., Skakkebaek, N. E., ... Toppari, J. (2007). Insulin-like factor 3 levels in cord blood and serum from children: Effects of age, postnatal hypothalamic–pituitary–gonadal axis activation, and cryptorchidism. *Journal of Clinical Endocrinology and Metabolism*, 92(10), 4020–4027. <https://doi.org/10.1210/jc.2007-0974>
- Bergada, I., Milani, C., Bedecarras, P., Andreone, L., Ropelato, M. G., Gottlieb, S., ... Rey, R. A. (2006). Time course of the serum gonadotropin surge, inhibins, and anti-Mullerian hormone in normal newborn males during the first month of life. *Journal of Clinical Endocrinology and Metabolism*, 91(10), 4092–4098. <https://doi.org/10.1210/jc.2006-1079>
- Berglund, A., Viuff, M. H., Skakkebaek, A., Chang, S., Stochholm, K., & Gravholt, C. H. (2019). Changes in the cohort composition of turner syndrome and severe non-diagnosis of Klinefelter, 47,XXX and 47,XYY syndrome: A nationwide cohort study. *Orphanet Journal of Rare Diseases*, 14(1), 16. <https://doi.org/10.1186/s13023-018-0976-2>
- Bianchi, D. W., & Wilkins-Haug, L. (2014). Integration of noninvasive DNA testing for aneuploidy into prenatal care: What has happened since the rubber met the road? *Clinical Chemistry*, 60(1), 78–87. <https://doi.org/10.1373/clinchem.2013.202663>
- Bin-Abbas, B., Conte, F. A., Grumbach, M. M., & Kaplan, S. L. (1999). Congenital hypogonadotropic hypogonadism and micropenis: Effect of testosterone treatment on adult penile size why sex reversal is not indicated. *Journal of Pediatrics*, 134(5), 579–583.
- Boas, M., Boisen, K. A., Virtanen, H. E., Kaleva, M., Suomi, A. M., Schmidt, I. M., ... Main, K. M. (2006). Postnatal penile length and growth rate correlate to serum testosterone levels: A longitudinal study of 1962 normal boys. *European Journal of Endocrinology*, 154(1), 125–129. <https://doi.org/10.1530/eje.1.02066>
- Bojesen, A., Juul, S., Birkebaek, N. H., & Gravholt, C. H. (2006). Morbidity in Klinefelter syndrome: A Danish register study based on hospital discharge diagnoses. *Journal of Clinical Endocrinology and Metabolism*, 91(4), 1254–1260. <https://doi.org/10.1210/jc.2005-0697>
- Bojesen, A., Juul, S., & Gravholt, C. H. (2003). Prenatal and postnatal prevalence of Klinefelter syndrome: A national registry study. *Journal of Clinical Endocrinology and Metabolism*, 88(2), 622–626. <https://doi.org/10.1210/jc.2002-021491>
- Bougneres, P., Francois, M., Pantalone, L., Rodrigue, D., Bouvattier, C., Demesteere, E., ... Lahlou, N. (2008). Effects of an early postnatal treatment of hypogonadotropic hypogonadism with a continuous subcutaneous infusion of recombinant follicle-stimulating hormone and luteinizing hormone. *Journal of Clinical Endocrinology and Metabolism*, 93(6), 2202–2205. <https://doi.org/10.1210/jc.2008-0121>
- Boukari, K., Meduri, G., Brailly-Tabard, S., Guibourdenche, J., Ciampi, M. L., Massin, N., ... Young, J. (2009). Lack of androgen receptor expression in Sertoli cells accounts for the absence of anti-Mullerian hormone repression during early human testis development. *Journal of Clinical Endocrinology and Metabolism*, 94(5), 1818–1825. <https://doi.org/10.1210/jc.2008-1909>
- Bouvattier, C., Carel, J. C., Lecoindre, C., David, A., Sultan, C., Bertrand, A. M., ... Chaussain, J. L. (2002). Postnatal changes of T, LH, and FSH in 46,XY infants with mutations in the AR gene. *Journal of Clinical Endocrinology and Metabolism*, 87(1), 29–32. <https://doi.org/10.1210/jcem.87.1.7923>
- Cabrol, S., Ross, J. L., Fennoy, I., Bouvattier, C., Roger, M., & Lahlou, N. (2011). Assessment of Leydig and Sertoli cell functions in infants with nonmosaic Klinefelter syndrome: Insulin-like peptide 3 levels are normal and positively correlated with LH levels. *Journal of Clinical Endocrinology and Metabolism*, 96(4), E746–E753. <https://doi.org/10.1210/jc.2010-2103>
- Clements, J. A., Reyes, F. I., Winter, J. S., & Faiman, C. (1976). Studies on human sexual development. III. Fetal pituitary and serum, and amniotic fluid concentrations of LH, CG, and FSH. *Journal of Clinical Endocrinology and Metabolism*, 42(1), 9–19. <https://doi.org/10.1210/jcem-42-1-9>
- Codesal, J., Regadera, J., Nistal, M., Regadera-Sejas, J., & Paniagua, R. (1990). Involution of human fetal Leydig cells. An immunohistochemical, ultrastructural and quantitative study. *Journal of Anatomy*, 172, 103–114.
- Coerd, W., Rehder, H., Gausmann, I., Johannisson, R., & Gropp, A. (1985). Quantitative histology of human fetal testes in chromosomal disease. *Pediatric Pathology*, 3(2–4), 245–259.
- Copeland, K. C., & Chernauek, S. (2016). Mini-puberty and growth. *Pediatrics*, 138(1), e20161301. <https://doi.org/10.1542/peds.2016-1301>
- Cortes, D., Muller, J., & Skakkebaek, N. E. (1987). Proliferation of Sertoli cells during development of the human testis assessed by stereological

- methods. *International Journal of Andrology*, 10(4), 589–596. <https://doi.org/10.1111/j.1365-2605.1987.tb00358.x>
- Davis, S. M., Kaar, J. L., Ringham, B. M., Hockett, C. W., Glueck, D. H., & Dabelea, D. (2019). Sex differences in infant body composition emerge in the first 5 months of life. *Journal of Pediatric Endocrinology and Metabolism*, 32(11), 1235–1239. <https://doi.org/10.1515/jpem-2019-0243>
- Davis, S. M., Reynolds, R. M., Dabelea, D. M., Zeitler, P. S., & Tartaglia, N. R. (2019). Testosterone treatment in infants with 47,XXY: Effects on body composition. *Journal of the Endocrine Society*, 3(12), 2276–2285. <https://doi.org/10.1210/js.2019-00274>
- de Mello, W. G., de Moraes, S. R., Dornelles, R. C., Kagohara Elias, L. L., Antunes-Rodrigues, J., & Bedran de Castro, J. C. (2012). Effects of neonatal castration and androgenization on sexual dimorphism in bone, leptin and corticosterone secretion. *Bone*, 50(4), 893–900. <https://doi.org/10.1016/j.bone.2011.12.009>
- Dkhal, M. A., Al-Quraishy, S., Abdel-Baki, A. A., Ghanjati, F., Arauzo-Bravo, M. J., Delic, D., ... Wunderlich, F. (2015). Epigenetic modifications of gene promoter DNA in the liver of adult female mice masculinized by testosterone. *Journal of Steroid Biochemistry and Molecular Biology*, 145, 121–130. <https://doi.org/10.1016/j.jsbmb.2014.11.006>
- Forest, M. G., De, P. E., & Bertrand, J. (1980). Testicular and adrenal androgens and their binding to plasma proteins in the perinatal period: Developmental patterns of plasma testosterone, 4-androstenedione, dehydroepiandrosterone and its sulfate in premature and small for date infants as compared with that of full-term infants. *Journal of Steroid Biochemistry*, 12, 25–36. [https://doi.org/10.1016/0022-4731\(80\)90247-2](https://doi.org/10.1016/0022-4731(80)90247-2)
- Forest, M. G., Sizonenko, P. C., Cathiard, A. M., & Bertrand, J. (1974). Hypophyso-gonadal function in humans during the first year of life. 1. Evidence for testicular activity in early infancy. *The Journal of Clinical Investigation*, 53(3), 819–828. <https://doi.org/10.1172/JCI107621>
- Ghahramani, N. M., Ngun, T. C., Chen, P. Y., Tian, Y., Krishnan, S., Muir, S., ... Vilain, E. (2014). The effects of perinatal testosterone exposure on the DNA methylome of the mouse brain are late-emerging. *Biology of Sex Difference*, 5, 8. <https://doi.org/10.1186/2042-6410-5-8>
- Gil, M. M., Accurti, V., Santacruz, B., Plana, M. N., & Nicolaides, K. H. (2017). Analysis of cell-free DNA in maternal blood in screening for aneuploidies: Updated meta-analysis. *Ultrasound in Obstetrics and Gynecology*, 50, 302–314. <https://doi.org/10.1002/uog.17484>
- Grumbach, M. M. (2005). A window of opportunity: The diagnosis of gonadotropin deficiency in the male infant. *Journal of Clinical Endocrinology and Metabolism*, 90(5), 3122–3127. <https://doi.org/10.1210/jc.2004-2465>
- Hatipoglu, N., & Kurtoglu, S. (2013). Micropenis: Etiology, diagnosis and treatment approaches. *Journal of Clinical Research in Pediatric Endocrinology*, 5(4), 217–223. <https://doi.org/10.4274/Jcrpe.1135>
- Hines, M., Spencer, D., Kung, K. T., Browne, W. V., Constantinescu, M., & Noorderhaven, R. M. (2016). The early postnatal period, mini-puberty, provides a window on the role of testosterone in human neurobehavioural development. *Current Opinion in Neurobiology*, 38, 69–73. <https://doi.org/10.1016/j.conb.2016.02.008>
- Howard-Bath, A., Poulton, A., Halliday, J., & Hui, L. (2018). Population-based trends in the prenatal diagnosis of sex chromosome aneuploidy before and after non-invasive prenatal testing. *Prenatal Diagnosis*, 38(13), 1062–1068. <https://doi.org/10.1002/pd.5363>
- Johannsen, T. H., Main, K. M., Ljubicic, M. L., Jensen, T. K., Andersen, H. R., Andersen, M. S., ... Juul, A. (2018). Sex differences in reproductive hormones during mini-puberty in infants with Normal and disordered sex development. *Journal of Clinical Endocrinology and Metabolism*, 103(8), 3028–3037. <https://doi.org/10.1210/jc.2018-00482>
- Kaiserman, K. B., Nakamoto, J. M., Geffner, M. E., & McCabe, E. R. (1998). Minipuberty of infancy and adolescent pubertal function in adrenal hypoplasia congenita. *Journal of Pediatrics*, 133(2), 300–302. [https://doi.org/10.1016/s0022-3476\(98\)70242-2](https://doi.org/10.1016/s0022-3476(98)70242-2)
- Kaplan, S. L., & Grumbach, M. M. (1976). The ontogenesis of human foetal hormones. II. Luteinizing hormone (LH) and follicle stimulating hormone (FSH). *Acta Endocrinologica (Copenhagen)*, 81(4), 808–829. <https://doi.org/10.1530/acta.0.0810808>
- Kiviranta, P., Kuiri-Hanninen, T., Saari, A., Lamidi, M. L., Dunkel, L., & Sankilampi, U. (2016). Transient postnatal gonadal activation and growth velocity in infancy. *Pediatrics*, 138(1), e20153561. <https://doi.org/10.1542/peds.2015-3561>
- Kuiri-Hanninen, T., Koskeniemi, J., Dunkel, L., Toppari, J., & Sankilampi, U. (2019). Postnatal testicular activity in healthy boys and boys with cryptorchidism. *Frontiers in Endocrinology (Lausanne)*, 10, 489. <https://doi.org/10.3389/fendo.2019.00489>
- Kuiri-Hanninen, T., Sankilampi, U., & Dunkel, L. (2014). Activation of the hypothalamic-pituitary-gonadal axis in infancy: Minipuberty. *Hormone Research in Pediatrics*, 82(2), 73–80. <https://doi.org/10.1159/000362414>
- Kuiri-Hanninen, T., Seuri, R., Tyrvalinen, E., Turpeinen, U., Hamalainen, E., Stenman, U. H., ... Sankilampi, U. (2011). Increased activity of the hypothalamic-pituitary-testicular axis in infancy results in increased androgen action in premature boys. *Journal of Clinical Endocrinology and Metabolism*, 96(1), 98–105. <https://doi.org/10.1210/jc.2010-1359>
- Kung, K. T., Browne, W. V., Constantinescu, M., Noorderhaven, R. M., & Hines, M. (2016). Early postnatal testosterone predicts sex-related differences in early expressive vocabulary. *Psychoneuroendocrinology*, 68, 111–116. <https://doi.org/10.1016/j.psyneuen.2016.03.001>
- Lahlou, N., Fennoy, I., Carel, J. C., & Roger, M. (2004). Inhibin B and anti-Müllerian hormone, but not testosterone levels, are normal in infants with nonmosaic Klinefelter syndrome. *Journal of Clinical Endocrinology and Metabolism*, 89(4), 1864–1868. <https://doi.org/10.1210/jc.2003-031624>
- Lahlou, N., Fennoy, I., Ross, J. L., Bouvattier, C., & Roger, M. (2011). Clinical and hormonal status of infants with nonmosaic XXY karyotype. *Acta Paediatrica*, 100(6), 824–829. <https://doi.org/10.1111/j.1651-2227.2011.02280.x>
- Lamminmaki, A., Hines, M., Kuiri-Hanninen, T., Kilpelainen, L., Dunkel, L., & Sankilampi, U. (2012). Testosterone measured in infancy predicts subsequent sex-typed behavior in boys and in girls. *Hormones and Behaviour*, 61(4), 611–616. <https://doi.org/10.1016/j.yhbeh.2012.02.013>
- Lanciotti, L., Cofini, M., Leonardi, A., Penta, L., & Esposito, S. (2018). Up-to-date review about minipuberty and overview on hypothalamic-pituitary-gonadal axis activation in fetal and neonatal life. *Frontiers in Endocrinology (Lausanne)*, 9, 410. <https://doi.org/10.3389/fendo.2018.00410>
- Lee, Y. S., Cheng, A. W., Ahmed, S. F., Shaw, N. J., & Hughes, I. A. (2007). Genital anomalies in Klinefelter's syndrome. *Hormone Research*, 68(3), 150–155. <https://doi.org/10.1159/000106375>
- Main, K. M., Schmidt, I. M., & Skakkebaek, N. E. (2000). A possible role for reproductive hormones in newborn boys: Progressive hypogonadism without the postnatal testosterone peak. *Journal of Clinical Endocrinology and Metabolism*, 85(12), 4905–4907. <https://doi.org/10.1210/jcem.85.12.7058>
- Main, K. M., Schmidt, I. M., Toppari, J., & Skakkebaek, N. E. (2002). Early postnatal treatment of hypogonadotropic hypogonadism with recombinant human FSH and LH. *European Journal of Endocrinology*, 146(1), 75–79. <https://doi.org/10.1530/eje.0.1460075>
- Main, K. M., Toppari, J., Suomi, A. M., Kaleva, M., Chellakooty, M., Schmidt, I. M., ... Skakkebaek, N. E. (2006). Larger testes and higher inhibin B levels in Finnish than in Danish newborn boys. *Journal of Clinical Endocrinology and Metabolism*, 91(7), 2732–2737. <https://doi.org/10.1210/jc.2005-2443>
- Massa, G., de Zegher, F., & Vanderschueren-Lodeweyckx, M. (1992). Serum levels of immunoreactive inhibin, FSH, and LH in human infants

- at preterm and term birth. *Biology of the Neonate*, 61(3), 150–155. <https://doi.org/10.1159/000243737>
- Muller, J., & Skakkebaek, N. E. (1984). Fluctuations in the number of germ cells during the late foetal and early postnatal periods in boys. *Acta Endocrinologica (Copenhagen)*, 105(2), 271–274. <https://doi.org/10.1530/acta.0.1050271>
- Murken, J. D., Stengel-Rutkowski, S., Walther, J. U., Westenfelder, S. R., Remberger, K. H., & Zimmer, F. (1974). Letter: Klinefelter's syndrome in a fetus. *Lancet*, 2(7873), 171. [https://doi.org/10.1016/s0140-6736\(74\)91608-0](https://doi.org/10.1016/s0140-6736(74)91608-0)
- Nugent, B. M., Wright, C. L., Shetty, A. C., Hodes, G. E., Lenz, K. M., Mahurkar, A., ... McCarthy, M. M. (2015). Brain feminization requires active repression of masculinization via DNA methylation. *Nature Neuroscience*, 18(5), 690–697. <https://doi.org/10.1038/nn.3988>
- Pasterski, V., Acerini, C. L., Dunger, D. B., Ong, K. K., Hughes, I. A., Thankamony, A., & Hines, M. (2015). Postnatal penile growth concurrent with mini-puberty predicts later sex-typed play behavior: Evidence for neurobehavioral effects of the postnatal androgen surge in typically developing boys. *Hormones and Behavior*, 69, 98–105. <https://doi.org/10.1016/j.yhbeh.2015.01.002>
- Ratcliffe, S. G. (1982). The sexual development of boys with the chromosome constitution 47,XXY (Klinefelter's syndrome). *Clinics in Endocrinology and Metabolism*, 11(3), 703–716. [https://doi.org/10.1016/s0300-595x\(82\)80008-x](https://doi.org/10.1016/s0300-595x(82)80008-x)
- Ratcliffe, S. G., Murray, L., & Teague, P. (1986). Edinburgh study of growth and development of children with sex chromosome abnormalities. III. *Birth Defects Original Article Series*, 22(3), 73–118.
- Robinson, A., Bender, B. G., & Linden, M. G. (1990). Summary of clinical findings in children and young adults with sex chromosome anomalies. *Birth Defects Original Article Series*, 26(4), 225–228.
- Ross, J. L., Samango-Sprouse, C., Lahlou, N., Kowal, K., Elder, F. F., & Zinn, A. (2005). Early androgen deficiency in infants and young boys with 47,XXY Klinefelter syndrome. *Hormone Research*, 64(1), 39–45. <https://doi.org/10.1159/000087313>
- Samango-Sprouse, C., Lasutschinkow, P., Powell, S., Sadeghin, T., & Gropman, A. (2019). The incidence of anxiety symptoms in boys with 47,XXY (Klinefelter syndrome) and the possible impact of timing of diagnosis and hormonal replacement therapy. *American Journal of Medical Genetics A*, 179(3), 423–428. <https://doi.org/10.1002/ajmg.a.61038>
- Samango-Sprouse, C., Stapleton, E. J., Lawson, P., Mitchell, F., Sadeghin, T., Powell, S., & Gropman, A. L. (2015). Positive effects of early androgen therapy on the behavioral phenotype of boys with 47,XXY. *American Journal of Medical Genetics, Part C: Seminars in Medical Genetics*, 169(2), 150–157. <https://doi.org/10.1002/ajmg.c.31437>
- Samango-Sprouse, C. A., Sadeghin, T., Mitchell, F. L., Dixon, T., Stapleton, E., Kingery, M., & Gropman, A. L. (2013). Positive effects of short course androgen therapy on the neurodevelopmental outcome in boys with 47,XXY syndrome at 36 and 72 months of age. *American Journal of Medical Genetics, Part A*, 161A(3), 501–508. <https://doi.org/10.1002/ajmg.a.35769>
- Schmidt, H., & Schwarz, H. P. (2000). Serum concentrations of LH and FSH in the healthy newborn. *European Journal of Endocrinology*, 143(2), 213–215. <https://doi.org/10.1530/eje.0.1430213>
- Stoupa, A., Samara-Boustani, D., Flechtner, I., Pinto, G., Jourdon, I., Gonzalez-Briceno, L., ... Beltrand, J. (2017). Efficacy and safety of continuous subcutaneous infusion of recombinant human gonadotropins for congenital micropenis during early infancy. *Hormone Research in Pediatrics*, 87(2), 103–110. <https://doi.org/10.1159/000454861>
- Swee, D. S., & Quinton, R. (2019). Congenital Hypogonadotrophic hypogonadism: Minipuberty and the case for neonatal diagnosis. *Frontiers in Endocrinology (Lausanne)*, 10, 97. <https://doi.org/10.3389/fendo.2019.00097>
- Swift-Gallant, A., Coome, L. A., Ramzan, F., & Monks, D. A. (2016). Non-neural androgen receptors affect sexual differentiation of brain and behavior. *Endocrinology*, 157(2), 788–798. <https://doi.org/10.1210/en.2015-1355>
- Tartaglia, N., Boada, C., Tanda, T., Brown, M., Crawford, L., Alston, A., ... Ross, J. (2020). Early neurodevelopmental and medical profile in children with sex chromosome trisomies: Background for the prospective eXtraordinary Babies Study to identify early risk factors and targets for intervention. *American Journal of Medical Genetics Part C: Seminars in Medical Genetics*. <https://doi.org/10.1002/ajmg.c.31788>
- Varimo, T., Hero, M., Laitinen, E. M., Miettinen, P. J., Tommiska, J., Kansakoski, J., ... Raivio, T. (2016). Childhood growth in boys with congenital hypogonadotropic hypogonadism. *Pediatric Research*, 79(5), 705–709. <https://doi.org/10.1038/pr.2015.278>
- Winter, J. S. (1982). Hypothalamic-pituitary function in the fetus and infant. *Clinics in Endocrinology and Metabolism*, 11(1), 41–55. [https://doi.org/10.1016/s0300-595x\(82\)80037-6](https://doi.org/10.1016/s0300-595x(82)80037-6)
- Winter, J. S., Faiman, C., Hobson, W. C., Prasad, A. V., & Reyes, F. I. (1975). Pituitary-gonadal relations in infancy. I. Patterns of serum gonadotropin concentrations from birth to four years of age in man and chimpanzee. *Journal of Clinical Endocrinology and Metabolism*, 40(4), 545–551. <https://doi.org/10.1210/jcem-40-4-545>
- Winter, J. S., Hughes, I. A., Reyes, F. I., & Faiman, C. (1976). Pituitary-gonadal relations in infancy: 2. Patterns of serum gonadal steroid concentrations in man from birth to two years of age. *Journal of Clinical Endocrinology and Metabolism*, 42(4), 679–686. <https://doi.org/10.1210/jcem-42-4-679>
- Zeger, M. P., Zinn, A. R., Lahlou, N., Ramos, P., Kowal, K., Samango-Sprouse, C., ... Ross, J. L. (2008). Effect of ascertainment and genetic features on the phenotype of Klinefelter syndrome. *Journal of Pediatrics*, 152(5), 716–722. <https://doi.org/10.1016/j.jpeds.2007.10.019>
- Zinn, A. R., Ramos, P., Elder, F. F., Kowal, K., Samango-Sprouse, C., & Ross, J. L. (2005). Androgen receptor CAGn repeat length influences phenotype of 47,XXY (Klinefelter) syndrome. *Journal of Clinical Endocrinology and Metabolism*, 90, 5041–5046. <https://doi.org/10.1210/jc.2005-0432>

How to cite this article: Aksglaede L, Davis SM, Ross JL, Juul A. Minipuberty in Klinefelter syndrome: Current status and future directions. *Am J Med Genet Part C*. 2020;184C: 320–326. <https://doi.org/10.1002/ajmg.c.31794>