Genetics inMedicine

www.nature.com/gim

Check for updates

ARTICLE

The effect of early hormonal treatment (EHT) on expressive and receptive language capabilities in boys with 47,XXY (Klinefelter syndrome) during infancy and early childhood

Carole Samango-Sprouse[1,2,3]✉, Michaela Reiko Brooks[3], Patricia Lasutchinkow[3], Teresa Sadeghin[3], Sherida Powell[4], Mary Pat Hamzik[3], Sophia Song[3] and Andrea L. Gropman[5,6]

PURPOSE: 47,XXY is associated with variable neurodevelopmental outcomes including deficits in expressive and receptive language development. Early hormonal treatment (EHT) has been associated with mitigating some deficiencies in boys with 47,XXY. This study investigates these language capabilities of 47,XXY boys in the first five years of life and the associated effects of EHT on these capabilities.

METHODS: One hundred and seventy-five boys with 47,XXY between the ages of 0 and 5 years, 11 months completed neurodevelopmental assessments specific to age examining their expressive and receptive language capabilities. Subjects were grouped by treatment (EHT and No-T) and differences were analyzed.

RESULTS: In the age groups of under 12 months, 24–35 months, 36–47 months, and 60–71 months, the EHT group scored significantly higher on expressive language assessments than the No-T group ($p = 0.09$, $p = 0.0002$, $p = 0.009$, and $p = 0.02$, respectively). In the age groups of under 12 months and 24–35 months, the EHT group scored significantly better on the auditory comprehension domain of the PLS-4/5 ($p = 0.02$ and $p = 0.05$, respectively) than the No-T group.

CONCLUSION: Study data suggest EHT may be essential in optimizing receptive and expressive language development in 47,XXY boys during early childhood, which is critical in fostering reading skills and later academic success.

Genetics in Medicine (2021) 23:1017–1022; https://doi.org/10.1038/s41436-021-01098-w

INTRODUCTION

47,XXY, or Klinefelter syndrome, is the most common sex chromosome aneuploidy (SCA), occurring in about 1 of every 650 male births.[1,2] The disorder, initially described in 1942, is characterized by small testicular size, tall stature with eunochoid body type, and aspermatogenesis.[3] Since then, androgen deficiency has been found to be a hallmark symptom of this disorder and a possible contributor to the neurodevelopmental deficiencies associated with 47,XXY.[4–6]

The neurodevelopmental profile of males with 47,XXY has been documented, and while there is wide variability in the phenotype, often language-based learning disabilities, motor apraxia, speech and language abnormalities, reading disorders, and neuromotor dysfunction are present.[5,7–10] Speech and language is one of the most impacted domains of 47,XXY, with approximately 80% of boys experiencing difficulties in these skills.[11,12] During infancy and early childhood, speech and language delays in this disorder are very common, with their expressive ability being almost always more impaired than their receptive skills.[13,14] A study conducted by Graham et al. in 1988 showed consistently lower verbal intelligence quotients (VIQ) in boys with 47,XXY compared with controls, while the performance intelligence quotients (PIQ) remained about equal or were minimally impacted.[14]

Early studies on young children with 47,XXY report lower performances in word retrieval, syntactic production, auditory processing and memory, and narrative formation.[14,15] Other previous notable studies have identified diminished abilities in phonological processes, oromotor skills, articulation, and motor planning of speech in 47,XXY.[7,11,16,17] Men with 47,XXY have been shown to have more challenges in discriminating emotional content during verbal tasks, and even more so, in tone of voice. These men made significantly more errors in the affective prosody condition (including loudness, timbre, intonation, etc.) than the semantics condition, though they showed deficits in both.[18] More recent studies, such as one conducted by van Rijn et al. in 2014, report two-thirds of boys with 47,XXY have poor social interactions and communication skills in early childhood although boys diagnosed prenatally were not segregated from those with postnatal diagnoses.[19] These early language deficits have been shown to precede difficulties in academics, particularly in reading and writing (dyslexia and dysgraphia),[16,20] as well as contribute as a risk factor for social anxiety, social isolation, and low self-esteem.[21,22]

In the last 15 years, there has been in increasing focus on early hormonal treatment (EHT) in boys with 47,XXY in which testosterone is administered during a "mini-puberty" in infancy to address their androgen deficiency (three injections of testosterone enanthate, 25 mg, each). This mini-puberty is believed to occur about two weeks after birth and continue until at least 24 weeks,[23,24] and is known to have a profound impact on brain development, masculinization of the infant boys, and promotion of social behaviors.[25,26] EHT in young boys with 47, XXY has been shown to be associated with mitigation of some of

[1]Department of Pediatrics, George Washington University, Washington, DC, USA. [2]Department of Human and Molecular Genetics, Florida International University, Miami, FL, USA. [3]Department of Research, The Focus Foundation, Davidsonville, MD, USA. [4]Department of Economics, George Washington University, Washington, DC, USA. [5]Department of Neurology, George Washington University, Washington, DC, USA. [6]Division of Neurogenetics and Developmental Pediatrics, Children's National Health System, Washington, DC, USA. ✉email: cssprouse@email.gwu.edu

© The Author(s), under exclusive licence to the American College of Medical Genetics and Genomics 2021

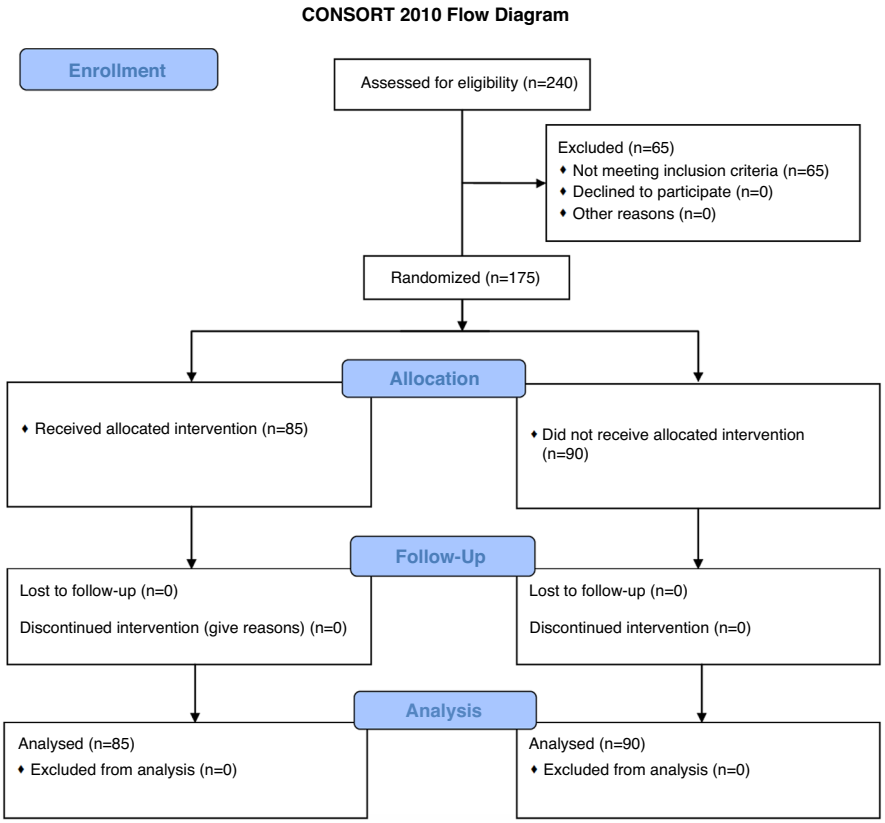

Fig. 1 Participant enrollment and follow-up. A total of 175 prenatally diagnosed infants and children with 47,XXY were included for analysis in this study (No-T total $N = 90$, T total $N = 85$).

the physical, cognitive, and behavioral impairments related to the disorder.[6,27,28]

While several studies have demonstrated the positive effect associated with androgen treatment in adolescents and adults, there are still few studies illustrating the potential influence of testosterone on infants with 47,XXY on their early expressive and receptive language skills. This study aims to investigate the possible positive association of early hormonal treatment on expressive and receptive language capabilities in boys with 47, XXY during the first five years of life.

MATERIALS AND METHODS

Subjects

Two hundred forty boys with the karyotype of 47,XXY between the ages of 1 month and 71 months were referred by their physicians, parents, and/or ancillary health-care providers throughout the country for a comprehensive neurodevelopmental assessment at the Neurodevelopmental Diagnostic Center for Children in Davidsonville, Maryland. The respective diagnoses were confirmed through karyotype analysis and documented in the children's medical records. The subjects included children with a prenatal diagnosis only, with exclusion criteria of postnatally diagnosed boys and the presence of copy-number variants (CNVs). None of the boys with 47,XXY in either group had neonatal seizures, significant birth complications requiring more than 3-day hospitalizations, or any other brain abnormalities. Of that initial group of 240 boys with 47, XXY, 175 qualified for the investigations and completed the required testing (Fig. 1).

The Focus Foundation, a nonprofit foundation, provided scholarship monies for those families in need to obtain neurodevelopmental assessments to minimize ascertainment bias. Pediatric endocrinologists throughout the country evaluated infants with 47,XXY and 85 (48.6%) infants received testosterone injections as a course of treatment and 90 (51.4%) did not. The timing and dosage was assessed on an individual basis, and necessary laboratory tests were determined by the pediatric endocrinologists and were often not obtained prior to neurodevelopmental testing.

Neurodevelopmental testing

Comprehensive neurodevelopmental testing was administered and selected based on the subject's chronological age. For this study, a neurodevelopmentalist (ND) was first trained to administer all assessments and, to ensure study fidelity, established 95% reliability in all language assessments with a licensed speech and language pathologist (SLP). The ND then trained all other testers individually for more than two weeks on all assessments according to specific guidelines outlined in each assessment's manual, and supervised the administration of these tests on numerous neurotypical children, as well as children with various genetic disorders. Reliability (95%) was achieved with these testers through observation, and reviewing test scores of more than ten neurotypical children prior to the administration of any assessments with study participants. Throughout the study, the ND randomly and routinely observed testing on subjects in both (EHT and No-T) groups. Testers (including the SLP) were blinded to the treatment status of each subject at the time of evaluation and assessments were randomly assigned to minimize researcher bias.

The language portion of Bayley Scales of Infant and Toddler Development 3rd/4th Edition was utilized to assess speech and language development in children from birth to 42 months of age. The Early Language Milestone Scale 2 (ELM-2) evaluates speech and language skills from birth to 3 years of age. The Preschool Language Scale (PLS) 4 or 5 was used to determine auditory comprehension and expressive communication ability for subjects from birth to 5 years, 11 months. The Verbal Intelligence Quotient on the Wechsler Preschool & Primary Scale of Intelligence 3rd Edition and Verbal Comprehension Index on the Wechsler Preschool & Primary Scale of Intelligence 4th Edition (WPPSI-III VIQ or WPPSI-IV VCI) was used to evaluate verbal reasoning, semantic knowledge, and verbal comprehension and expression in subjects from 2 years, 6 months to 5 years, 11 months. The Receptive One Word Picture Vocabulary Test–Revised (ROWPVT-R/4) and Expressive One Word Picture

Table 1. Neurodevelopmental assessments.

Preschool Language Scale 4/5 (PLS-4/5) • Auditory Comprehension • Verbal Ability/Expressive Communication
Early Language Milestone Scale 2 (ELM-2) • Expressive Ability • Receptive Ability
Bayley Scales of Infant and Toddler Development 3/4 • Language Scale
Expressive One Word Picture Vocabulary Test–Revised (EOWPVT-R/4)
Receptive One Word Picture Vocabulary Test–Revised (ROWPVT-R/4)
Wechsler Preschool & Primary Scale of Intelligence-III/IV (WPPSI-III/IV) • Verbal IQ/Verbal Comprehension Index (VCI)
Child Behavior Checklist (CBCL-Lang) • Language Domain
MacArthur–Bates Communicative Development Inventories • Words and Gestures (CDI-WG) • Words and Sentences (CDI-WS) • Vocabulary Checklist (CDI-VC)

Vocabulary Test–Revised (EOWPVT-R/4) were utilized to evaluate receptive and expressive vocabulary skills in children beginning at 2 years of age (Table 1).

Not all children completed all assessments due to fatigue, which is a common occurrence when evaluating infants and young children. Some children were lost to follow-up for a variety of reasons such as relocation or compliance. If multiple scores for an individual patient on a given subtest were available, all test scores falling within the appropriate age range for the subtest were used in the analyses.

Behavioral forms were completed by primary caregivers on the child's speech and language capabilities with the Child Behavior Checklist-Language Scale (CBCL-Lang), MacArthur–Bates Communicative Development Inventories (CDI): Words and Gestures (CDI-WG), Words and Sentences (CDI-WS), and Vocabulary Checklist (CDI-VC).

Statistical analysis

The cohort was separated into six groups based on chronological age: under 12 months, 12 to 23 months, 24 to 35 months, 36 to 47 months, 48 to 59 months, and 60 to 71 months. Neurodevelopmental data for each age group were then further bifurcated into two cohorts for analysis based on those infants who received EHT (EHT group) and those untreated (No-T group).

The skewness–kurtosis test was used to assess normality of the groups, then a test of significance was performed using the two-sample Wilcoxon rank-sum test when normality was present and the Mann–Whitney test if normality was absent. The null hypothesis is that there is no statistically significant difference between the means of the group 1 (treated) and group 2 (untreated) on any developmental domain or subtest.

To minimize confounding factors, subjects were evaluated by examiners who were blinded to who had received early hormonal therapy. A data coordinator scored all standardized assessments and was unaware of the child's treatment status. A program manager randomly reviewed charts and data entry to ensure that all scores were consistent from testing protocols, maintaining fidelity of the study process from data entry to data analysis. A biostatistician completed the data analysis offsite and did not directly interact with any patients.

RESULTS

Demographics of the EHT group and No-T group are similar including maternal age, delivery, and parental education levels (Table 2). The majority of subjects were Caucasian in each group with one African American and seven Hispanics in the EHT group and two African Americans and seven Hispanics in the No-T group. The No-T group weighed an average of 3.32 kg at birth, while the EHT group weighed an average of 3.26 kg; the two groups were not significantly different.

Table 2. Parental patient characteristics and demographics of the study population ($N = 175$).

47,XXY ($N = 175$)			
Patient background		Parental background	
EHT group ($N = 85$)			
Mean birth weight	3.26 kg (7.19 lbs)	Mean maternal age	36.99
Delivery	Vaginal, 44	Mean paternal age	38.40
	Cesarean, 36	Mean maternal education	5.47
Race	Caucasian, 45	Mean paternal education	5.45
	Hispanic, 7		
	African American, 1		
	Other, 6		
No-T group ($N = 90$)			
Mean birth weight	3.32 kg (7.32 lbs)	Mean maternal age	37.45
Delivery	Vaginal, 46	Mean paternal age	37.96
	Cesarean, 28	Mean maternal education	5.35
Race	Caucasian, 49	Mean paternal education	4.98
	Hispanic, 7		
	African American, 2		
	Other, 6		

EHT early hormonal treatment, *No-T* no treatment.

A positive treatment effect on multiple stages of development at the $P = 0.05$ level was associated with the EHT. In the under 12 months group, the auditory comprehension subtests revealed significance at the $P = 0.02$ level for those in the EHT group (Table 3). In expressive communication, there was a trend toward significance for the EHT group at $P = 0.08$. On the Bayley language assessment, the EHT group was significantly better than the No-T group ($P = 0.04$).

In the 12 to 23 month group, on the Bayley language assessment, there was a significant difference between the EHT group and the No-T group ($P = 0.05$). In the 24 to 35 month group, the PLS revealed greater significance between those boys in the EHT group versus the No-T group on both the auditory comprehension subtest and expressive communication subtest ($P = 0.05$ and $P = 0.0002$, respectively) (Table 4). On the ELM expressive language subtest ($P = 0.03$), EOWPVT-R/4 ($P = 0.05$), and Bayley language ($P = 0.05$), the EHT group scored significantly higher than the No-T group.

The EOWPVT-4 revealed significance in the 36 to 47 month group at $P = 0.01$ (Table 3). In contrast, there was a trend toward significance between the EHT compared with No-T on the ROWPVT-4 in the same group.

There were no significant findings for the 48 to 59 month group on any of the speech and language assessments. However, in the 60 to 71 month age group, there was a significant difference between the two groups at $P = 0.02$ on the expressive

Table 3. Neurodevelopmental results in age groups <12 months, 12–23 months, 36–47 months, 48–59 months, and 60–71 months.

Age group	Subtest	EHT Group			No-T Group			*P* value
		N	Mean	Median	*N*	Mean	Median	
<12 months	PLS-4/5 Auditory comprehension	51	100.04	100	50	95.38	93	0.02
	PLS-4/5 Expressive communication	51	99.92	99	50	95.18	91	0.09[a]
	Bayley language 3/4	33	106.25	108.5	20	101.1	100	0.05
12–23 months	Bayley language 3/4	43	108.26	106	6	97.5	97	0.05
36–47 months	EOWPVT-R/4	25	116	119	25	105.47	102	0.009
60–71 months	PLS-4/5 Expressive communication	16	108.88	112	13	96.85	93	0.02
	WPPSI-III/IV Verbal IQ	15	111.93	112.5	16	101	104	0.6[a]

EHT early hormonal treatment, *EOWPVT-R/4* Expressive One Word Picture Vocabulary Test–Revised, *No-T* no treatment, *PLS-4/5* Preschool Language Scale 4/5, *WPPSI-III/IV* Wechsler Preschool & Primary Scale of Intelligence 3rd/4th Edition.
[a]$P = 0.1$ level.

Table 4. Neurodevelopmental results in 24–35 month age group.

Test type	EHT group			No-T Group			*P* value
	N	Mean	Median	*N*	Mean	Median	
PLS-4/5 Auditory comprehension	39	105.84	103.5	33	99.94	98	0.05
PLS-4/5 Expressive communication	38	105.03	106	33	95.36	91	0.0002
ELM-2 Expressive language	30	101.83	100	18	94.05	91	0.03
EOWPVT-R/4	30	105.4	106.5	25	100.32	100	0.05
ROWPVT-R/4	30	107.28	109	28	102.61	99.5	0.09[a]
Bayley Language Scale 3/4	22	109.32	109	4	97.25	98.5	0.05
CDI-WS Complexity	12	13.75	15.50	17	4.82	1	0.02
CDI-WS Mean Length	16	4.08	3.67	17	2.23	2.33	0.08[a]
CBCL Combines Words	3	0.83	1	23	0.33	0	0.06[a]

CBCL Child Behavior Checklist, *EHT* early hormonal treatment, *ELM-2* Early Language Milestone Scale 2, *EOWPVT-R/4* Expressive One Word Picture Vocabulary Test–Revised, *No-T* no treatment, *PLS-4/5* Preschool Language Scale 4/5, *ROWPVT-R/4* Receptive One Word Picture Vocabulary Test–Revised, *WPPSI-III/IV* Wechsler Preschool & Primary Scale of Intelligence 3rd/4th Edition, *CDI-WS* Words and Sentences.
[a]$P = 0.1$ level.

communication subtest. There was a trend toward significance between the EHT group on the WPPSI verbal intelligence scale ($P = 0.06$).

On the CDI-WS, in the 24 to 35 month age group, there was significant difference between the EHT group and the No-T group on the complexity of speech patterns at the $P = 0.02$ level and trend toward potential significance on mean length ($P = 0.08$) (Table 4). On the CBCL, the EHT group had significantly more combinations of words on the Combines Words scale than the No-T group ($P = 0.06$).

DISCUSSION

Our current study in infants and young boys with 47,XXY suggests a positive association between early hormonal treatment and the potential mitigation of early expressive and receptive language deficits. Our findings suggest that an early course of testosterone is associated with improved function in auditory comprehension, expressive ability, and verbal intelligence. This provides further support for the link between neurobiological treatment and improved neurodevelopmental outcome in boys with XXY.[5,6,9,28–30]

We observed a positive effect of treatment in auditory comprehension for the groups of under 12 months and between 24 to 35 months. A research study completed by Benasich and Tallal in 2002 found that early deficits in rapid auditory processing abilities both precede and predict subsequent language delays.[31] Our findings of improved auditory comprehension in the EHT groups within these ages suggest that testosterone may provide a mechanism to foster improved speech development or conversely may be a preventive measure for the early and common expressive language delays seen in 47,XXY boys. Since the early language dysfunction is an indicator of later language-based learning disabilities, EHT may be impactful on one of the most common phenotypic features of 47,XXY.

Our findings of delayed expressive language capabilities in the untreated group are consistent with several previous studies in older 47,XXY boys and men.[9,14,27,29] However, this innovative study identifies these expressive language delays in a large group of prenatally diagnosed infants and toddlers with 47,XXY. Furthermore, the EHT group had no expressive language delays, which provides a possible pathway to minimize, if not eliminate, these expressive differences prior to the associated social

interaction deficits and low self-esteem development. Though not a focus of this current study, St. John et al. recently described speech capabilities in boys with 47,XXY from infancy to adolescence.[16] Future studies should expand on this aspect of the phenotypic presentation by exploring the role of EHT on speech articulation, production, and phonological processing in boys with 47,XXY.

The discrepancy between receptive and expressive ability in 47, XXY is not well understood although it is a common and pervasive aspect of this disorder. 47,XXY is commonly associated with low muscle tone, which may lead to impaired oral motor skills. A recently published paper by our group reported that there was increased incidence of difficulties with latching on during feeding in the first few months of life, which is associated with decreased muscle tonus in the oral motor musculature.[30] Decreased oral muscle tonus contributes to reduced control of the mouth and throat muscles, thereby negatively impacting speaking ability and reflecting lower cognitive performance than true cognitive level.[8,9,32] A number of earlier studies have demonstrated that the majority of two-year-olds with poor early expressive language skills have a lasting impact in and continue to show these deficits until at least four years of age,[33,34] and testosterone is known to impact the muscle strength and muscle tonus in these boys.[5,10] EHT may indirectly improve expressive language capabilities by enhancing oral motor skills in infants with 47,XXY, and subsequently foster a more appropriate developmental track commensurate with neurotypical children.

The 60 to 71 month age group is the only group in which the untreated boys showed significantly lower verbal intelligence that those with EHT. It is well recognized that speech and language dysfunction in the early years of life is highly associated with later various school challenges. Therefore, this significant difference between the two groups may be likely attributed to the beginning of academic difficulty children have when exhibiting language delay in the preschool years.[35–37] Difficulties in language development may become more salient as well as impactful in the school setting where children are learning to read and write. Our observations suggest EHT may be associated with minimizing this expressive and selected aspects of receptive language delays and lower verbal intelligence in later development of 47,XXY boys.

In this study, the 24 to 35 month age group showed significance associated with EHT across multiple language assessments. This is not surprising, as there is a well-documented language explosion that occurs from 18 to 26 months of age in which infants substantially broaden their vocabulary and increase word production.[38,39] This age group is therefore where we would expect (and found) the expressive delay in untreated 47,XXY toddlers, and age appropriate expressive and receptive language skills in the EHT toddlers, to be most evident. This early and appropriate language explosion may be a later predictor of the higher verbal IQ and decreased academic deficits in 47,XXY.

There are some limitations of this study. The CBCL and MacArthur surveys may be prone to bias due to the parent-reported measures. However, in our study, the parental measures were not the only findings documenting the specific language findings in 47,XXY. In many studies on 47,XXY there is a predisposition to ascertainment bias since 75% of individuals are not identified in their lifetime. However, our study provides the largest study to date of those children prenatally diagnosed with 47,XXY. Additionally, the various confounding factors that are common to many studies have been minimized whenever possible. The differences between the EHT group and the No-T group such as timing of diagnosis, socioeconomic status, and parental education were homogenized to maximize our understanding of the relationship between neurobiological treatment and neurodevelopmental outcome.

This study provides additional evidence of positive neurobiological treatment effects within the first five years of life for 47,XXY boys, which is important for several reasons. Our findings of improved neurodevelopmental trajectory in association with EHT treated infants with XXY further elucidate the value of early detection and treatment. Our study showed no adverse side effects in any of our infants at the time of the study. Equally compelling, the oldest participants in previous EHT studies are now approaching the end of adolescence and young adulthood, and there have been no adverse effects, which is quite encouraging that this is both safe and effective in addressing some of the neurodevelopmental issues associated with 47,XXY.

With more infants with 47,XXY being identified prenatally due to noninvasive prenatal screening (NIPS), there is considerable opportunity for earlier treatment and fostering optimal outcome for these boys. Additionally, the benefits of testosterone treatment appear to be quite timely and during such a critical developmental period, our findings suggest that the appropriate development of expressive and receptive language capabilities is essential, as it sets the foundation for success in later social settings and academics. Continued research is needed to determine the optimal timing of treatment, as well as the effect of EHT on other aspects of neurodevelopment in infants with 47, XXY.

DATA AVAILABILITY

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Received: 24 September 2020; Revised: 4 January 2021; Accepted: 4 January 2021;
Published online: 24 February 2021

REFERENCES

1. Bojesen, A., Juul, S. & Gravholt, C. H. Prenatal and postnatal prevalence of Klinefelter syndrome: a national registry study. *J. Clin. Endocrinol. Metab.* **88**, 622–626 (2003).
2. Morris, J. K., Alberman, E., Scott, C. & Jacobs, P. Is the prevalence of Klinefelter syndrome increasing? *Eur. J. Hum. Genet.* **16**, 163–170 (2008).
3. Klinefelter, H. F. Jr., Reifenstein, E. C. Jr. & Albright, F. Jr. Syndrome characterized by gynecomastia, aspermatogenesis without A-Leydigism, and increased excretion of follicle-stimulating hormone. *J. Clin. Endocrinol.* **2**, 615–667. (1942).
4. Lahlou, N., Fennoy, I., Carel, J. C. & Roger, M. Inhibin B and anti-Mullerian hormone, but not testosterone levels, are normal in infants with nonmosaic Klinefelter syndrome. *J. Clin. Endocrinol. Metab.* **89**, 1864–1868 (2004).
5. Ross, J. L., Samango-Sprouse, C., Lahlou, N., Kowal, K., Elder, F. F. & Zinn, A. Early androgen deficiency in infants and young boys with 47, XXY Klinefelter syndrome. *Horm. Res. Paediatr.* **64**, 39–45 (2005).
6. Samango-Sprouse, C. A. et al. Positive effects of short course androgen therapy on the neurodevelopmental outcome in boys with 47, XXY syndrome at 36 and 72 months of age. *Am. J. Med. Genet. A* **161**, 501–508 (2013).
7. Samango-Sprouse, C. & Rogol, A. XXY: the hidden disability and a prototype for an infantile presentation of developmental dyspraxia (IDD). *Infants Young Child.* **15**, 11–18 (2002).
8. Simpson, J. L., Elias, S. Sex chromosomal polysomies (47, XXY; 47, XYY; 47, XXX), sex reversed (46, XX) males, and disorders of the male reproductive ducts. *Genet Obstet Gynacol. Philadelphia: Saunders*. 323–341 (2003).
9. Ross, J. L., Zeger, M. P., Kushner, H., Zinn, A. R. & Roeltgen, D. P. An extra X or Y chromosome: contrasting the cognitive and motor phenotypes in childhood in boys with 47, XYY syndrome or 47, XXY Klinefelter syndrome. *Ment. Retard. Dev. Disabil. Res. Rev.* **15**, 309–317 (2009).
10. Zeger, M. P. et al. Effect of ascertainment and genetic features on the phenotype of Klinefelter syndrome. *J. Pediatr.* **152**, 716–722 (2008).
11. Bender, B., Fry, E., Pennington, B., Puck, M., Salbenblatt, J. & Robinson, A. Speech and language development in 41 children with sex chromosome anomalies. *Pediatrics.* **71**, 262–267 (1983).

12. Ratcliffe, S. G. 6 The sexual development of boys with the chromosome constitution 47, XXY (Klinefelter's syndrome). *Clin. Endocrinol. Metab.* **11**, 703–716 (1982).
13. Samango-Sprouse, C. Mental development in polysomy X Klinefelter syndrome (47, XXY; 48, XXXY): effects of incomplete X inactivation. *Semin. Reprod. Med.* **19**, 193–202 (2001).
14. Graham, J. M., Bashir, A. S., Stark, R. E., Silbert, A. & Walzer, S. Oral and written language abilities of XXY boys: implications for anticipatory guidance. *Pediatrics.* **81**, 795–806 (1988).
15. Walzer, S., Bashir, A. S. & Silbert, A. R. Cognitive and behavioral factors in the learning disabilities of 47, XXY and 47, XYY boys. *Birth Defects Orig. Artic. Ser.* **26**, 45 (1990).
16. St John, M. et al. Speech and language in children with Klinefelter syndrome. *J. Commun. Disord.* **78**, 84–96 (2019).
17. Walzer, S. et al. A method for the longitudinal study of behavioral development in infants and children: the early development of XXY children. *J. Child Psychol. Psychiatry* **19**, 213–229 (1978).
18. Van Rijn, S., Aleman, A., Swaab, H., Krijn, T., Vingerhoets, G. & Kahn, R. S. What it is said versus how it is said: comprehension of affective prosody in men with Klinefelter (47, XXY) syndrome. *J. Int. Neuropsychol. Soc.* **13**, 1065 (2007).
19. van Rijn, S., Barendse, M., van Goozen, S. & Swaab, H. Social attention, affective arousal and empathy in men with Klinefelter syndrome (47, XXY): evidence from eyetracking and skin conductance. *PLoS One* **9**, e84721 (2014).
20. Peterson, R. L., Pennington, B. F., Shriberg, L. D. & Boada, R. What influences literacy outcome in children with speech sound disorder? *J. Speech Lang. Hear. Res.* **52**, 1175–1188 (2009).
21. Samango-Sprouse, C. Expansion of the phenotypic profile of the young child with XXY. *Pediatr. Endocrinol. Rev.* **8**, 160–168 (2010).
22. van Rijn, S., de Sonneville, L. & Swaab, H. The nature of social cognitive deficits in children and adults with Klinefelter syndrome (47, XXY). *Genes Brain Behav*. **17**, e12465 (2018).
23. Andersson, A. M. et al. Longitudinal reproductive hormone profiles in infants: peak of inhibin B levels in infant boys exceeds levels in adult men. *J. Clin. Endocrinol. Metab.* **83**, 675–681 (1998).
24. Forest, M. G., Sizonenko, P. C., Cathiard, A. M. & Bertrand, J. Hypophyso-gonadal function in humans during the first year of life: I. Evidence for testicular activity in early infancy. *J. Clin. Invest.* **53**, 819–828 (1974).
25. Nielsen, J., Sørensen, A. M. & Sørensen, K. Mental development of unselected children with sex chromosome abnormalities. *Hum. Genet.* **59**, 324–332 (1981).
26. Lewis, K. & Lee, P. A. Endocrinology of male puberty. *Curr. Opin. Endocrinol. Diabetes Obes.* **16**, 5–9 (2009).
27. Lanfranco, F., Kamischke, A., Zitzmann, M. & Nieschlag, E. Klinefelter's syndrome. *Lancet* **364**, 273–283 (2004).
28. Samango-Sprouse, C. et al. Positive effects of early androgen therapy on the behavioral phenotype of boys with 47, XXY. *Am. J. Med. Genet. C* **169**, 150–157 (2015).
29. Mandoki, M. W., Sumner, G. S., Hoffman, R. P. & Riconda, D. L. A review of Klinefelter's syndrome in children and adolescents. *J. Am. Acad. Child Adolesc. Psychiatry* **30**, 167–172 (1991).
30. Samango-Sprouse, C., Yu, C., Porter, G., Tipton, L., Lasutschinkow, P., Sadeghin, T. A review of the intriguing interaction between testosterone and neurocognitive development in males with 47,XXY. *Curr. Opin. Obstet. Gynecol.* (in press).
31. Benasich, A. A. & Tallal, P. Infant discrimination of rapid auditory cues predicts later language impairment. *Behav. Brain Res.* **136**, 31–49 (2002).
32. Salbenblatt, J. A., Meyers, D. C., Bender, B. G., Linden, M. G. & Robinson, A. Gross and fine motor development in 47, XXY and 47, XYY males. *Pediatrics.* **80**, 240–244 (1987).
33. Rescorla, L. & Schwartz, E. Outcome of toddlers with specific expressive language delay. *Appl. Psycholinguist.* **11**, 393–407 (1990).
34. Paul, R. & Smith, R. L. Narrative skills in 4-year-olds with normal, impaired, and late-developing language. *J. Speech Lang. Hear. Res.* **36**, 592–598 (1993).
35. Aram, D. M., Ekelman, B. L. & Nation, J. E. Preschoolers with language disorders: 10 years later. *J. Speech Lang. Hear. Res.* **27**, 232–244 (1984).
36. Garvey, M. & Gordon, N. A follow-up study of children with disorders of speech development. *Br. J. Disord. Commun.* **8**, 17–28 (1973).
37. McLaughlin, M. R. Speech and language delay in children. *Am. Fam. Physician* **83**, 1183–1188 (2011).
38. Goldfield, B. A. & Reznick, J. S. Early lexical acquisition: rate, content, and the vocabulary spurt. *J. Child Lang.* **17**, 171–183 (1990).
39. Huttenlocher, J., Haight, W., Bryk, A., Seltzer, M. & Lyons, T. Early vocabulary growth: relation to language input and gender. *Dev. Psychol.* **27**, 236 (1991).

ACKNOWLEDGEMENTS

We would like to express our deep gratitude to all our participants and their families. We would like to thank The Focus Foundation for their continued funding of our investigative studies.

AUTHOR CONTRIBUTIONS

Conceptualization: C.S.-S., A.L.G. Data curation: M.R.B., S.P., T.S. Formal analysis: S.P. Funding acquisition: C.S.-S. Investigation: C.S.-S., M.R.B., P.L. Methodology: C.S.-S., M.R.B., P.L., A.L.G. Project administration: C.S.-S., A.L.G. Resources: C.S.-S., A.L.G. Supervision: C.S.-S., A.L.G. Validation: C.S.-S., M.R.B., P.L., A.L.G. Visualization: C.S.-S.,M.R.B. Writing—original draft: C.S.-S., M.R.B., P.L. Writing—review & editing: C.S.-S., M.R.B., S. S., M.P.H, T.S., A.L.G.

ETHICS DECLARATION

Parents consented for their children to receive the comprehensive neurodevelopmental evaluation. The Western Institutional Review Board (WIRB) was approved for this study protocol (20081226), informed consent was completed on each study participant, and all participant data have been de-identified. This study adheres to the principles outlined in the Declaration of Helsinki.

COMPETING INTERESTS

The authors declare no competing interests.

ADDITIONAL INFORMATION

Correspondence and requests for materials should be addressed to C.S.-S.

Reprints and permission information is available at http://www.nature.com/reprints

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.