SPECTRUM OF XY DISORDERS OF SEX DEVELOPMENT IN PAKISTAN

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ABSTRACT

Objective: To determine the clinico-endocrinal spectrum of XY Disorders of Sex Development (DSD) according to the new classification in our population.

Study Design: Cross sectional study.

Place and Duration of Study: Department of Chemical Pathology and Endocrinology Armed Forces Institute of Pathology Rawalpindi, from Jan 2012 to Aug 2015.

Material and Methods: A total of 151 patients who reported for work-up of DSD during 2012-2015 and labeled XY on karyotyping were included in the study. Patients without karyotyping results, mixed sex chromosome or XX karyotypes were excluded from the study. Patient's clinical features and results of serum hormones i.e. LH, FSH, Testosterone, 17 hydroxy progesterone, DHEAS and hCG stimulation test were analyzed.

Results: Out of 151 patients of XY DSD, 68 (45%) patients were diagnosed as partial androgen insensitivity syndrome (PIAS), 18 (12%) as isolated micropenis, 25 (16.6%) as partial gonadal dysgenesis, 12 (7.9%) as hypogonadotropic hypogonadism, 11 (7.2%) as primary hypogonadism, 13 (8.6%) as complete androgen insensitivity syndrome (CAIS) and 4 (2.6%) as 5α reductase deficiency. There was a wide variation in age of presentation ranging from three months to twenty five years with the median age of 9.5 years. Nineteen (12.6%) were raised as female and 132 (87.4%) were raised as male. The main complaints at presentation were ambiguous genitalia (30.5%), undescended testes (21.2%), delayed puberty (19.2%), micropenis (16.6%), inguinal hernia (8.6%), and primary amenorrhea (3.9%).

Conclusion: Androgen insensitivity syndrome (AIS) in its various forms is the commonest of all XY DSD. All DSD including AIS require complete investigations in a tertiary care setup.

Keywords: Common clinical features, Etiological distribution, XY DSD (XY Disorders of Sex Development).

INTRODUCTION

Disorders of sex development (DSD) are defined as congenital conditions in which development of chromosomal, gonadal, or anatomical sex is atypical[1]. In 2006 an umbrella term 'DSD' was proposed by the Lawson Wilkins Pediatric Endocrine Society (LWPES) and the European Society for Paediatric Endocrinology (ESPE). They also proposed a new etiological classification on the basis of karyotype analysis[2]. Although the term DSD is generally accepted by medical professionals but still some patients and support groups do not accept this term and stigma of disorder[3,4]. The disorders of sex development are now broadly grouped into three main diagnostic categories namely 46 XY DSD (previously called male Pseudo hermaphroditism), 46 XX DSD (previously called female Pseudo hermaphroditism) and sex chromosome DSD (previously called true hermaphroditism). The sex chromosome DSD also include 45X/46XY mixed gonadal dysgenesis, Turner's syndrome (TS) and Klinefelter's syndrome (KS)[5-7].

Among 46 XY individuals the overall reported incidence of DSD is 1 in 20,000 births[8]. The frequency of ovotesticular DSDs is 1:100,000 live births and that of testicular or mixed gonadal dysgenesis is 1:10,000[9]. However when all congenital anomalies are considered like cryptorchidism and hypospadias the incidence is much higher i.e. 1:200 to 1:300[10]. But presently the diagnosis of specific DSD is generally limited to only those with proximal hypospadias along with

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cryptorchidism. These reported estimates provide a useful perspective.

A general approach to investigation of DSD includes karyotyping and locating gonads by palpation, ultrasound or other imaging techniques. 46, XY DSD can be due to several etiologies and require a more extensive diagnostic evaluation. These disorders can result from gonadal dysgenesis due to mutations in one of the several genes that are involved in testicular development[11]. Disorders of androgen synthesis include mutations in genes in the pathway of testosterone synthesis that either are common to the adrenal gland and the testes or are just found in the testes. These are all autosomal recessive conditions. Androgen insensitivity syndrome is due to mutations in the androgen receptor on Xq11, and thus is an X-linked recessive condition[12]. Congenital hypogonadotropic hypogonadism can be due to one of several gene defects[13,14]. Fourty Six XY DSD have multiple genetic as well as non-genetic etiologies. However exact etiology cannot be assigned in 30-40% of cases[15]. Foury Six XY DSD is frequently noticed in children with reduced prenatal growth. But no other associated malformation or steroidogenic defect is detected in such children. Other studies have shown that around 30% of undetermined 46 XY DSD cases are associated with low birth weight, indicating that adverse events in early pregnancy are frequent causes of congenital non-genetic 46, XY DSD[16,17].

Present study was conducted with the objectives to find the etiological distribution of XY DSD according to new classification and determine the most commonly encountered clinical features among our XY DSD patients.

PATIENTS AND METHODS

This cross sectional study was conducted in the Department of Chemical Pathology and Endocrinology, Armed Forces Institute of Pathology, Rawalpindi (AFIP) from January 2012 to August 2015, after approval of Ethical Review Committee, AFIP Rawalpindi and gate keeper`s permission. Informed consents of the patients or parents were also obtained. A total of 151 cases (sample size calculated using WHO sample size calculator) of DSD were included by non-probability consecutive sampling. The patients with clinical presentation of DSD who were labeled XY on karyotyping were included in the study, whereas patients without karyotyping results, mixed sex chromosome or XX Karyotypes were excluded from the study. The details of clinical features included age at presentation, main complaints, assigned gender and family history. Criteria suggesting DSD included ambiguous genitalia (i.e. apparent female

Table-I: Age distribution.

Age (Years)	Frequency	Percentage (%)
<1	11	7.3
1-13	99	65.6
13-18	35	23.2
18-25	6	4.0
Total	151	100.0

Table-II: Presenting complaints of XY DSD patients.

Presenting complaint	Frequency	Percentage (%)
Ambiguous genitalia	46	30.5
Delayed puberty	29	19.2
Primary amenorrhoea	6	3.9
Undescended testes	32	21.2
Micropenis	25	16.6
Inguinal hernia	13	8.6
Total	151	100.0

genitalia with clitoromegaly, posterior labial fusion or inguinal/labial mass and apparent male genitalia with non-palpable testes), micropenis, undescended testis (testes presenting as inguinal hernia or abdominal testes found on imaging studies), incomplete or delayed puberty and primary amenorrhea.

Patient's clinical features and results of Serum hormones i.e. LH, FSH, Testosterone, 17 hydroxy progesterone, DHEAS and hCG stimulation test were analyzed. LH, FSH, testosterone were measured by immunoassay using Beckman Coulter commercial kits on DXI 800 Analyzer. 17-OH Progesterone was measured by Enzyme linked immune-Sorbent assay using Demeditec commercial kits. DHEAS was measured by chemiluminescence immune assay using commercial kits by Siemens on Immulite 2000. The hCG stimulation test was performed to detect the presence of functioning testicular tissue. Basal serum samples were collected for testosterone, FSH and LH, followed by I/M injection hCG 100 IU/Kg. Sample for serum testosterone was collected after 72 hours (on third day). Two to nine fold rise in serum testosterone was considered as adequate (normal) response, less than two fold rise was considered inadequate response and more than ten folds rise was considered as exaggerated response.

The under virilized males who had normal/raised basal LH, FSH, testosterone but adequate response to hCG stimulation were diagnosed as cases of AIS. Among these ones with normal female external genitalia were considered as complete AIS (CAIS) and rest as partial AIS (PAIS). While those showing exaggerated testosterone response to hCG stimulation were considered as 5α- reductase deficiency[18]. Primary hypogonadism was diagnosed in patients with high basal FSH, LH, low testosterone and no response on hCG stimulation. While those with high basal FSH, LH, low testosterone but low normal response to hCG stimulation were diagnosed as partial gonadal dysgenesis[19]. Hypo-gonadotropic hypogonadism was diagnosed in patients with low FSH, LH, testosterone and inadequate response to hCG stimulation[19]. Idiopathic micropenis was diagnosed in patients of normal male genitalia with micropenis, normal FSH, LH, testosterone and adequate response on hCG stimulation[19].

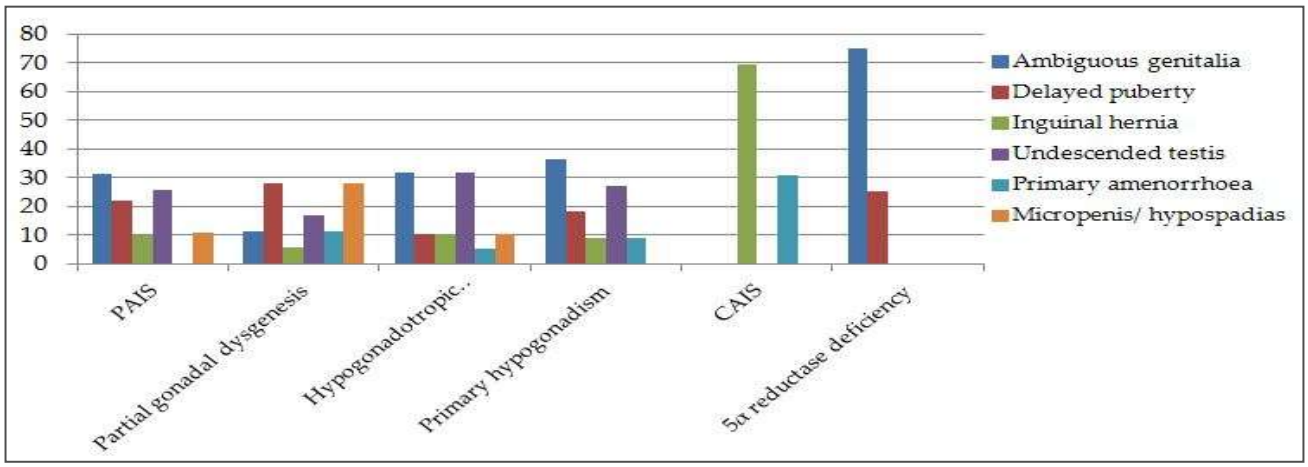

Figure: Presenting complaints of patients with XY DSD according to etiological diagnosis.

The data was analyzed using SPSS version 20. Mean ± SD was calculated for age, serum LH, FSH and testosterone levels. Frequencies and percentages were determined for age distribution, gender, presenting complaints, etiological diagnosis and HCG stimulation test.

RESULTS

Overall 151 patients fulfilled the criteria of XY DSD. The age at presentation was quite variable ranging from three months to twenty

five years with mean age of 9 ± 5.9 years. Eleven patients presented in infancy, ninety nine between 1 and 13 years, thirty five between 13 and 18 years and only six above 18 years of age (table-I). At the time of presentation 19 (12.6%) patients were raised as female and 132 (87.4%) as male. The XY DSD patients mainly presented with ambiguous genitalia, delayed puberty, primary amenorrhea, undescended testes, micro penis and inguinal mass as shown in table-II. Figure shows the distribution of complaints with different etiological diagnosis. Although ambiguous genetalia was the commonest presentation but CAIS mainly presented with inguinal hernia or primary amenorrhea and partial gonadal dysgenesis presented with delayed puberty and micropenis or hypospadias.

HCG stimulation test showed adequate testosterone response in 99 (65.6%), inadequate response in 48 (31.8%) and exaggerated response in 4 (2.6%). Presumptive diagnosis of PAIS, isolated micro penis, partial gonadal dysgenesis, hypo gonadotropic hypogonadism, primary hypogonadism, CAIS and 5-α reductase deficiency was made as shown in table-III. Mean value of serum luteinizing hormone was 2.74 ± 1.38 m IU/ml in PAIS, 25.8 ± 9.92 m IU/ml in partial gonadal dysgenesis, 0.31 ± 0.23 m IU/ml in hypo gonadotropic hypogonadism, 31.9 ± 13.07 m IU/ml in primary hypogonadism, 5.2 ± 1.45 m IU/ml in CAIS and 7 ± 2.2 m IU/ml in 5-α reductase deficiency. Whereas the mean value of serum testosterone was 1.20 ± 0.74 nmol/l in PAIS, 0.85 ± 0.65 nmol/l in partial gonadal dysgenesis, 0.51 ± 0.42 nmol/l in hypo gonadotropic hypogonadism, 0.63 ± .04 nmol/l in primary hypogonadism, 1.37 ± 0.97 nmol/l in CAIS and 1.70 ± 0.85 nmol/l in 5-α reductase deficiency. Biochemical profile & dynamic function test response in various etiological diagnosis of XY DSD are summarized in table-IV.

Table-III: Etiological diagnosis of XY DSD.

Etiological diagnosis	Frequency	Percentage (%)
PAIS	68	45
Isolated micropenis	18	12
Partial gonadal dysgenesis	25	16.6
Hypogonadotropic hypogonadism	12	7.9
Primary hypogonadism	11	7.3
CIAS	13	8.6
5-reductase deficiency	4	2.6

Table-IV: Biochemical profile & dynamic function test response in various etiological diagnosis of XY DSD.

Etiological diagnosis	Biochemical profile (LH, FSH, Testosterone)	Dynamic test response (HCG-stimulation)
PAIS	Normal/raised	Adequate
Isolated micropenis	Normal	Adequate
Partial gonadal dysgenesis	Raised LH/FSH Low Testosterone	Low normal
Hypo-gonadotropic hypogonadism	Low LH/FSH Low Testosterone	Inadequate
Primary hypogonadism	Raised LH/FSH Low Testosterone	No response
CIAS	Normal/raised	Adequate
5-reductase deficiency	Normal/raised	Exaggerated

DISCUSSION

The group of Disorders of Sex Development (DSD) with 46XY karyotype can be due to several etiologies and require more extensive diagnostic evaluation. They are broadly divided

into three main groups: 1) disorders of gonadal development, 2) disorders of testosterone synthesis and 3) disorders of testosterone action. The incidence of 46XY DSD is estimated at 1 in 20,000 live male births[8]. In one study 52% of all DSD patients had 46 XY DSD. Overall in 46 XY DSD patients a definitive diagnosis is made in less than 50% of patients and among diagnosed cases partial androgen insensitivity syndrome is the most common cause[20]. To our knowledge there is no study in our set up regarding evaluation of clinical features and etiological classification of XY DSD as per new classification system of DSD published in 2006[2]. We believe to gather some interesting data which can help in evaluation and diagnosis of XY DSD as per new system.

Majority of the DSD cases are picked up in newborn period with ambiguous genital development. However an individual can present later in childhood, adolescence or adulthood. These later presentations include progressive clitoromegaly, inguinal/labial mass, delayed or incomplete puberty, and progressive pubertal virillization in a phenotypic female or cyclical hematuria in a phenotypic male. Consanguinity among parents is a helpful clue to autosomal recessive disorders.

The age of presentation and gender assignment in XY DSD are variable depending on clinical presentation, cultural, socio-economic and educational factors. In this study majority (65.6%) presented between the age of 1 and 13 years. Mostly they were reared as males (87.4%). This in general indicates preference for male gender in our society. These findings are similar to another Pakistani study by Atta et al[21]. However in study by Erdogan et al mostly patients presented less than 1 year of age. As far as sex of rearing was concerned 39% were not assigned any gender before complete work up, 38% were assigned female gender and 23% male gender[18]. This may be due to better awareness among parents to seek medical advice and a more systematic approach towards gender assignment in DSD cases.

The patients with XY DSD present with different clinical manifestations. The commonest being ambiguous genetalia[18,21] followed by undescended testes, delayed puberty, micro penis, inguinal hernia and primary amenorrhea.

The data in this study demonstrates that the commonest cause of XY DSD is androgen insensitivity syndrome, PAIS (45%) being much more common than CAIS (8.6%). This is in concordance with previous studies[18,21-23]. However the percentage of PAIS patients in this study is higher than these studies. Molecular genetic analysis is required to confirm mutations in genes present in X, Y or autosomal chromosomes causing disorders of sex differentiation. Other etiological diagnosis of our XY DSD patients included hypo gonadotropic hypogonadism (7.9%), primary hypogonadism (7.3%), 5 alpha reductase deficiency (2.6%), partial gonadal dysgenesis (16.6%), isolated micro penis (12%) which is similar to studies by Atta et al and Erdogan et al[18,21].

The work up of DSD needs a very systematic approach. After detailed history and examination, the current approach is to (a) identify the sex chromosome complement by karyotype analysis or FISH with X and Y probes and chromosome microarray; (b) gather additional phenotypic information by complete metabolic and endocrine testing as well as imaging studies, and (c) genetic testing for copy number variants in regions associated with known DSD genes or gene sequencing either for single candidate gene or a gene panel. Biochemical profiles include 17-hydroxyprogesterone, serum electrolytes, androgen, anti-Mullerian hormone (AMH), gonadotropin levels, hCG and ACTH stimulation tests. Our study had limitation of not being able to carry out genetic testing and AMH assay.

Mostly there is a trend to assign male gender to XY DSD patients. However every individual case needs to be assessed on the basis of androgen responsiveness and CNS androgen exposure during fetal life. Gender assignment is

based on physical development, hormone secretion, genetic assessment and response to hormone therapy especially DHT (dihydro-testosterone). Health professionals need to properly communicate with the parents and the affected individual; so as to develop clear understanding about DSDs and their specific diagnosis. Education and psychological support are needed for each individual to cope up with the condition, relate to their community and establish relationships. Like other chronic conditions DSDs when diagnosed in pediatric years need lifelong follow up and transition to adult care. Persons with DSDs may not, in short term, experience negative consequences from poor treatment compliance or avoidance of care providers. However, such avoidance places the person at risk of long-term complications like gonadal malignancy, osteoporosis and poor psychosexual and psychosocial adaptation. In our set up there is need for proper awareness and acceptance especially among parents to help manage such individuals.

CONCLUSION

Androgen insensitivity syndrome (AIS) in its various forms is the commonest of all XY DSD. All DSD including AIS require complete investigations in a tertiary care setup.

CONFLICT OF INTEREST

This study has no conflict of interest to declare by any author.

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