**ORIGINAL RESEARCH ARTICLE**

# Clinical and genetic characteristics of disorders of sex development in Sudanese patients

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## Abstract

Given the scarce data on DSD in Sudan, we aimed to characterize DSD's clinical and genetic profile in Sudanese patients. We studied 60 patients with DSD using clinical data, cytogenetics, and PCR for the SRY gene. The results showed that 65% grew up as females and 35% as males. There was a high percentage of consanguineous parents (85%). Female genital mutilation (FGM) was performed in 75% of females. Patients who presented after pubertal age were 63%, with ambiguous genitalia in 61.7%, followed by primary amenorrhea (PA) in 30%. The *SRY* gene was positive in 3.3% of patients with 46,XX karyotype and negative in 6.7% of patients with 46,XY karyotype. 5αR2D-DSD was seen in 43.3%, gonadal dysgenesis in 21.7%, Ovotesticular syndrome in 6.7%, Swyer and Turner syndrome in 5% each, and Androgen Insensitivity Syndrome (AIS) in 3.3%. In conclusion, DSD in Sudan has a distinct profile with late presentation, dominated by 5αR2D-DSD due to the increased consanguineous marriage, and FGM represents a significant risk for DSD patients. *(Afr J Reprod Health 2024; 28 [4]: 13-21).*

**Keywords:** DSD, Chromosomal analysis, SRY gene, FGM

## Résumé

Compte tenu du peu de données sur le DSD au Soudan, nous avons cherché à caractériser le profil clinique et génétique du DSD chez les patients soudanais. Nous avons étudié 60 patients atteints de DSD en utilisant des données cliniques, cytogénétiques et PCR pour le gène SRY. Les résultats ont montré que 65 % ont grandi en tant que femmes et 35 % en tant qu'hommes. Il y avait un pourcentage élevé de parents consanguins (85 %). Des mutilations génitales féminines (MGF) ont été pratiquées chez 75 % des femmes. Les patientes qui se sont présentées après l'âge pubertaire étaient 63 %, avec des organes génitaux ambigus dans 61,7 %, suivis d'une aménorrhée primaire (AP) dans 30 %. Le gène SRY était positif chez 3,3 % des patients de caryotype 46,XX et négatif chez 6,7 % des patients de caryotype 46,XY. Le 5αR2D-DSD a été observé dans 43,3 %, la dysgénésie gonadique dans 21,7 %, le syndrome ovotesticulaire dans 6,7 %, le syndrome de Swyer et Turner dans 5 % chacun et le syndrome d'insensibilité aux androgènes (AIS) dans 3,3 %. En conclusion, le DSD au Soudan présente un profil distinct avec une présentation tardive, dominé par le 5αR2D-DSD en raison de l'augmentation des mariages consanguins, et les MGF représentent un risque important pour les patients DSD. *(Afr J Reprod Health 2024; 28 [4]: 13-21).*

**Mots-clés**: DSD, Analyse chromosomique, gène SRY, MGF

## Introduction

Disorders of sex development (DSD) are a collection of congenital conditions that impact the natural progression of sex chromosomes, gonads, and physical sex characteristics[1]. Due to the complexity and diversity of DSD, the same genetic anomalies can lead to varied clinical outcomes. Worldwide, the incidence of DSD is estimated to occur in approximately 0.1 to 0.2% of live births, affecting 1 in every 4,500 to 5,500 births[2].

Although Several classification systems have been developed to offer a comprehensive overview of DSD, they still need to provide a definitive summary of the different forms of DSD due to the existing nomenclature designating the various disorders. However, a simple classification of DSDs includes 46,XX DSD, 46,XY DSD, Sex

chromosome DSD, XX Sex reversal, XY Sex reversal, and Ovotesticular disorder[3]. The identification of the SRY gene and its role in male sex differentiation by Sinclair in 1990 has provided valuable insights into DSD[4]. The SRY gene is critical in male sex differentiation and development since its mutation accounts for 10–15% of 46,XY DSD cases. However, the spectrum of SRY gene mutations and other autosomal gene roles has yet to be fully understood[5].

The diagnosis of DSD is considered if sex determination and differentiation are incomplete or if there is a discrepancy between the individual's genotype and phenotype sex[6]. However, accurate diagnosis is sometimes challenging, and the incidence rate may vary across different geographic regions, considering the inherited type of DSD in areas with high consanguineous marriage and high prevalence of female genital mutilation (FGM) that may mask the ambiguity of genitalia[7]. In Sudan, DSD case incidence and genetic characteristics are not thoroughly studied because of the diagnosis challenges encountered by patients in low healthcare facilities, e.g., the average cost of investigating a case ranged between 250 and 300$[8]. On top of that, the psychological pressures of DSD, including stigma and social marginalization, contribute to significant mental health issues, and it is common for parents to experience shame, anxiety, and sadness following their child's diagnosis[9]. This study aims to illustrate the epidemiology, etiology, and clinical characteristics of DSDs in Sudanese patients. The results of this study will provide valuable insights into the magnitude of DSD in Sudan.

# Methods

## *Patients*

The present study included 60 patients referred to Elite Clinic between 2007 and 2010 with provisional diagnoses of DSD. Each patient completed a pre-designed questionnaire to gather demographic data and clinical history. A physical examination was also performed to evaluate secondary sexual characteristics as well as the external genitalia. The Sudanese intersex working group (SIWG) protocol was followed for DSD diagnosis and management, which included cytogenetic and molecular analysis, a pelvic ultrasound (U/S), hormonal profile assessment for follicle-stimulating hormone (FSH), luteinizing hormone (LH), testosterone levels, and human chorionic gonadotropin (hCG) stimulation test.

## *Sample collection*

Each patient had 5 mL of venous blood collected. Out of the 5 mL, 3 mL were placed in a sterile container with sodium heparin for lymphocyte culture and chromosomal analysis within two hours. The remaining 2 mL were placed in an EDTA container for DNA extraction and SRY gene molecular analyses.

## *Cytogenetic analyses*

Blood culture and cell harvesting were performed as previously described. In brief, 10 drops of blood were added to 10 ml of McCoy's 5A medium with L-glutamine in a 25 ml sterile conical flask. The media was supplemented with 25% fetal bovine serum (Sigma®), 3.4% phytohemagglutinin (Sigma® 10 µg/ml), and 1% penicillin/streptomycin at a concentration of 10000 U. The culture incubator was set at 37°C and 5% CO2 for 72 hours. After this period, culture harvesting was initiated by adding 0.1 ml of 10 µg/ml Colcemid to arrest lymphocytes in metaphase. The sample was then centrifuged at 1000 rpm in a swing-out centrifuge for 10 minutes, after which the supernatant was removed. Cells were resuspended in 6-7 ml of KCL (Hypotonic solution) with a concentration of 0.075. The supernatant was again discarded, and cells were resuspended in fresh fixative (3:1). Samples were then centrifuged, and fixation was repeated at least 4 times. 1-4 drops of the cell suspension were placed on a clean, dry microscope slide, and 2-4 slides were prepared for each patient.

The prepared slides were aged overnight by incubating them in the oven at 65°C. The slides were then incubated in a water bath using a standard saline concentration solution (2XSSC) at 65°C for 2-3 hours. They were then washed thoroughly using distilled water and left to dry. The slides were then stained using Wright's stain. Five to twenty-five metaphases were analyzed using the CytoVision system, Applied Imaging®, and photographed. The clonality criteria and the karyotypic descriptions

were based on the recommendations of the International System for Chromosomal Nomenclature (ISCN) (2020)[10].

### *DNA extraction*

Genomic DNAs were extracted manually using the guanidine chloride method. In brief, the collected blood was transferred to a 15 ml falcon tube after adding 10 ml of red cell lysis buffer (RCLB). The sample was then vortexed and centrifuged for 5 minutes at 6000 rpm. The supernatant was discarded, and the sample was vortexed again. The previous step was repeated until a clear pellet of white blood cells appeared at the bottom of the tube. Next, 2 ml of white blood cell lysis buffer (WCLB), 20 µL of proteinase K (10mg/mL), 1 ml of Guanidine chloride, and 300 µL of ammonium acetate (7.5M) were added to the sample. The sample was then vortexed to achieve homogeneity. One of two alternatives was then applied: incubating at 37°C overnight after cooling the sample to room temperature or adding 2ml of pre-chilled chloroform and vortexing. The sample was centrifuged for 10 minutes at 6000 rpm. The upper layer of the sample was collected in a new tube, and 10 ml of cold absolute ethanol was added to it. The sample was kept at -20 °C for 2 hours, shaken, and then centrifuged at 12000 rpm for 10 minutes. The supernatant was again discarded after adding 2 ml of 70% concentration ethanol, and the sample was centrifuged for 7 minutes at 12000 rpm or 14 minutes at 6000 rpm. The supernatant was removed, and the pellet was allowed to dry. 100 µL of ddh2o was added to each sample. DNA samples were kept at 4°C overnight and then at -20 °C until they were used for PCR analysis.

### *Polymerase chain reaction (PCR)*

The SRY gene was amplified through PCR using DNA samples from patients, fertile XX females, and fertile XY males (who acted as controls). The amplifications were carried out as described before, with the help of the primers XES10 and XES11, which flank the SRY open reading frame of sequence pY53.3, generating a 778 bp fragment[11].

To conduct the amplifications, 50-100 ng of genomic DNA, 0.3µL of Taq polymerase (1.5 U), 0.5 µL of each primer, 2.0 µL of dNTPs (100 mM), 1.5 µL of MgCL2 (1.5mM), 2.5µL of PCR buffer(10X) were combined and filled to 25µL with dd $H_2O$. The reactions were initiated with a 2-minute incubation at 94°C, followed by 32 cycles of cycling for 80 seconds at 94°C (Denaturation), 1.5 minutes at 60°C (Annealing), and 2.5 minutes at 71°C (Extension) using an automated cycler (Techne TC-3000). The reaction products were separated on 1% agarose-TBE gels containing 0.5 µg/ml ethidium bromide, using 1X TBE buffer as running (electrophoresis) buffer. A voltage of 120 volts was applied to the gel, with a Hyper ladder 100 bp loaded with the samples, and reaction products were documented with a gel documentation system.

### *Statistical analysis*

Categorical variables were analyzed using SPSS 23 software, and descriptive statistics were presented as frequencies and percentages.

## Results

In this study, 60 patients with Disorders of Sexual Development (DSD) between the ages of one month and 45 years were enrolled. Of these patients, 38 (63.3%) sought treatment after reaching puberty, which is defined as 14 years for boys and 12 years for girls. The remaining 22 (36.7%) patients were accessible before puberty. According to their assigned sex, 39 (65%) patients were raised as females, while 21 (35%) were raised as males (Table 1).

According to the study's findings, the majority of patients, 70%, belonged to the Afro-Asiatic tribe, while 28.3% belonged to the Nilo-Saharan tribe, and only 1.7% belonged to the Niger-Congo tribe. Of the patients, 85% had a history of consanguineous marriage, while the remaining 15% did not. The patients' clinical presentations varied, with 61.7% presenting with ambiguous genitalia, 30% with primary amenorrhea, and the remaining 8.3% presenting with other complaints, such as infertility in 3 (5.0%) cases, impotence, and Gynecomastia, each presenting in one patient (Table 1).

The cytogenetic analyses showed 46,XY karyotypes in 37 (61.7%) patients, 46,XX karyotypes in 20 (33.3%) patients, and abnormal karyotype results in only 3 patients (5.0%). Of those, 2 (3.3%) patients had mosaic 46,XX/46,XY karyotype, and one patient (1.7%) had mosaic 46,XX/45,XO karyotype (Table 1).

**Table 1:** The clinical data (frequency and percentage) of 60 patients with DSD

| | | Frequency | Percentage % |
|---|---|---|---|
| **Age** | **Range** | **1 month – 45 years** | |
| | Before puberty | 22 | 36.7 |
| | After puberty | 38 | 63.3 |
| **Assigned sex** | Male | 21 | 35.0 |
| | Female | 39 | 65.0 |
| **Tribal origin** | Afro-Asiatic | 42 | 70.0 |
| | Nilo-Saharan | 17 | 28.3 |
| | Niger-Congo | 1 | 1.7 |
| **History of consanguineous marriage** | Yes | 51 | 85.0 |
| | No | 9 | 15.0 |
| **Presenting complaint** | Ambiguous genitalia | 37 | 61.7 |
| | Primary amenorrhea | 18 | 30.0 |
| | Infertility | 3 | 5.0 |
| | Impotence | 1 | 1.7 |
| | Gynecomastia | 1 | 1.7 |
| **Chromosomal analysis** | 46,XX | 20 | 33.3 |
| | 46,XY | 37 | 61.7 |
| | 46,XX/46,XY | 2 | 3.3 |
| | 46,XX/45,X | 1 | 1.7 |
| **PCR for SRY gene** | Positive | 36 | 60.0 |
| | Negative | 24 | 40.0 |

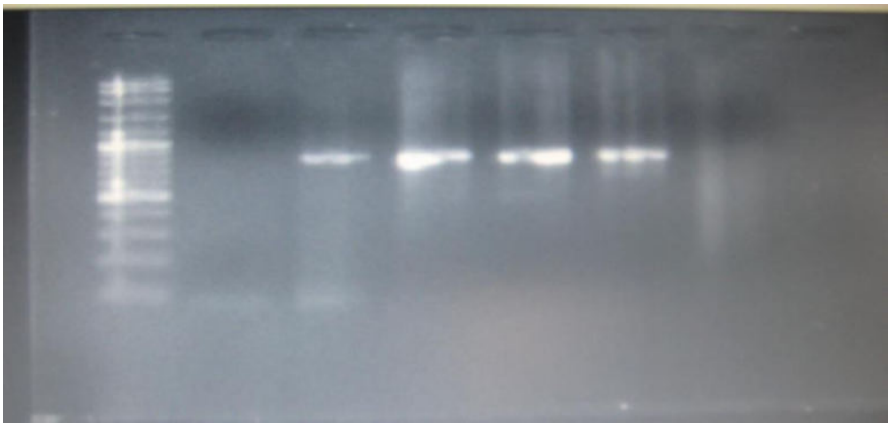

**Figure 1:** PCR amplification of the SRY gene from left to right: (1)100bp ladder, (2) -ve control (Fertile female), (3) +ve control (Fertile male), (lane 4-6) patients

**Table 2:** The final diagnoses in 60 patients with DSD disorders

| **Diagnosis** | | **Number (%)** |
|---|---|---|
| | 5 alpha reductase deficiencies | 26 (43.3) |
| 46,XY DSD 31 (51.7%) | Swyer syndrome | 3 (5.0) |
| | Androgen Insensitivity Syndrome (AIS) | 2 (3.3) |
| 46,XX DSD 18 (30%) | Complete Gonadal dysgenesis (CGD) | 13 (21.7) |
| | Turner syndrome (TS) | 3 (5.0) |
| | Gonadal agenesis | 2 (3.3) |
| OT DSD 4 (6.7) | Ovotesticular DSD | 4 (6.7) |
| | Syndromes with Ambiguous genitalia | 4 (6.7) |
| Others 7 (11.6%) | Infertility | 2 (3.3) |
| | Impotence | 1 (1.7) |
| **Total** | | 60 (100) |

**Table 3:** Summary of clinical data and final diagnoses in all 60 patients

| Age | History | Examination | Pelvic U/S | Hormonal | Karyotype | SRY gene | Final Diagnosis | Count (%) |
|---|---|---|---|---|---|---|---|---|
| Before or after puberty | Ambiguous genitalia, PA | Undescended testes, vagina with Clitoromegaly, circumcised | Short blind vagina, Absent uterus, bilateral undescended testes | ↔ FSH + LH ↑testosterone, +ve HCG stimulation test | 46XY | +ve | 5 alpha reductase deficiencies | 26 (43.3) |
| After puberty | PA | Underdeveloped SSC, normal female genitalia | Infantile uterus, small ovaries or not seen | ↑FSH and LH ↔ testosterone | 46XX | -ve | Gonadal dysgenesis | 13 (21.7) |
| Before or After puberty | Ambiguous genitalia | Unilateral Palpable gonads, ± big phallus and vagina | Small uterus, intra-abdominal gonad | ↔ FSH, LH , ↑ testosterone | 46XY 46XX 46XX/46XY | +ve | Ovotesticular DSD | 4 (6.7) |
| After puberty | PA | Female phenotype, Circumcised | Uterus and gonads present | ↑FSH and LH ↔ testosterone | 46XY | -ve | Swyer syndrome | 3 (5.0) |
| Before or After puberty | PA | short webbed neck, wide distance nipples | Infantile uterus, streakovaries | ↑FSH and LH | 46XX 45X, 46XX | -ve | Turner syndrome | 3 (5.0) |
| After puberty | PA | Normal SSC, short vagina | Absence of uterus, ovaries not seen | ↔ FSH, LH | 46XX | -ve | Gonadal agenesis | 2 (3.3) |
| After puberty | PA | Female phenotype, normal external genitalia or circumcised | Absent uterus, bilateral undescended testes | ↔ FSH, LH ↑ testosterone | 46XY | +ve | AIS | 2 (3.3) |
| Before puberty | Ambiguous genitalia | Dysmorphic features | Not done | Not done | 46XY 46XX | -ve / +ve | Syndromes with Ambiguous genitalia | 4 (6.7) |
| After puberty | Infertility | Male phenotype, normal male genitalia (penis and testes) | No uterus or ovaries were seen | ↔ FSH, LH, testosterone | 46XY | +ve | Infertility | 2 (3.3) |
| After puberty | Impotence | Male phenotype, normal male genitalia (penis and testes) | No uterus or ovaries were seen | ↔ FSH, LH, testosterone | 46XY | +ve | Impotency | 1 (1.7) |
| **Total** | | | | | | | | 60 (100) |

The study included 60 patients, of which 39 (65%) grew up as females during adulthood. Among these, 18 (30.0%) had a 46,XX karyotype, 20 (33.3%) had a 46,XY karyotype, and one (1.7%) was diagnosed with Turner syndrome and had a mosaic karyotype of 46,XX/45,X. On the other hand, 21 patients (33.3%) grew up as males during adulthood, with 17 (28.3%) having a 46,XY karyotype, 2 (3.3%) having a 46,XX karyotype, and the remaining 2 (3.3%) having a 46,XX/46,XY karyotype. Out of the 38 (63.3%)

patients who presented after puberty, 25 (41.7%) showed no secondary sexual characteristics (SSC), while 13 (21.7%) exhibited SSC.

Analysis using RCR of the SRY gene revealed that the gene was present in 36 patients (60%) (Figure 1). Among these, 31 (51.7%) had a 46,XY karyotype, 3 (5.0%) had a 46,XX karyotype, and 2 (3.3%) had a 46,XX/46,XY karyotype. On the other hand, the gene was absent in 24 patients (40%), with 17 (28.3%) having a 46,XX karyotype, 6 (10.0%) having a 46,XY karyotype, and 45,XO/46,XX karyotype (Table 1).

The patient's clinical presentation, physical examination, hormonal profiles, imaging studies, and cytogenetic and molecular analyses determined a provisional diagnosis of 46 XY DSD for 31 (51.7%) patients. Of these 31 patients, 26 (43.3%) were diagnosed with 5 alpha reductase deficiencies, 5 had rare familial 5 alpha reductase deficiency, 3 (5.0%) had Swyer syndrome, and 2 (3.3%) were diagnosed with Androgen Insensitivity Syndrome (AIS). However, 18 (30.0%) patients were diagnosed with 46,XX DSD, with 13 (21.7%) having complete gonadal dysgenesis (CGD), 3 (5.0%) having Turner syndrome, and only 2 (3.3%) having gonadal agenesis. Additionally, 4 (6.7%) patients were confirmed to have ovotesticular DSD. The remaining 7 (11.6%) patients had various other diagnoses, including 4 (6.7%) with syndromes with ambiguous genitalia, 2 (3.3%) with infertility, and only one patient with impotency (Tables 2 and 3).

## Discussion

The birth of a child with sexual anatomy that cannot be easily distinguished as a male or a female is a source of confusion and frustration to the parents, especially in countries with low health resources. In these countries, reports indicated that 64.8% of DSD patients were born attended by low-skilled midwives at home and that 52.2% of the cases are not observed at birth by the labor attendee, with a greater chance of wrong sex assignment of the newborn[12]. In the best scenario, if the ambiguity of the genitalia is recognized, the lobber attendee fails to deliver proper counseling, leaving parents in desolation and confusion. In such countries, patients and families used to suffer from stigmatization because of intense psychological pressure and social exclusion, leading to suicide in some cases[13]. On top of that comes the financial burden of the high cost of investigation, e.g., in Sudan, the average cost to investigate a case of DSD ranges between 250-300$ depending on the subtype of the DSD[8]. These include hormonal and enzymatic profiles, cytogenetic and molecular analysis, imaging studies, histopathology examination of gonadal biopsies, not to mention the cost of diagnostic and constructive surgery if needed. All these factors may delay the diagnosis process, leading to the loss of some patients before reaching the final diagnosis[14].

This particular study stands out as it sheds light on the relatively unexplored topic of Sudanese patients with disorders of sex development (DSD) who were diagnosed at a later stage. Late diagnosis indicates that these patients and their families may not have been aware of the condition until puberty. Nevertheless, previous reports have pointed to several factors contributing to the delayed diagnosis of DSD patients in developing countries, such as the incapacity of untrained healthcare providers to identify the issue, social stigma, financial constraints, lack of knowledge, reluctance to discuss the matter, and reliance on inexperienced practitioners[14].

The study shows that the most common sexual disorder is 46,XY DSD, accounting for 51.6%. This is followed by 46,XX DSD, which is seen in 30%, and OT DSD, which is seen in 6.7%. The most common type of all DSD in the study was 46,XY DSD due to 5α-Reductase 2 deficiency (5αR2D), accounting for 43.3% of all cases. It is worth noting that these results are different from those found in Europe, where congenital adrenal hyperplasia (CAH) is the most common type. This difference is due to the fact that in Europe, more than 95% of cases are diagnosed within the first few weeks of life, which includes those patients with CAH. In contrast, only 63.3% of our patients were diagnosed after puberty, with 30.0% complaining of primary amenorrhea as the main concern[15]. Nonetheless, our findings are not in line even with earlier research on DSD from Sudan, which demonstrated a comparable predominance of congenital adrenal hyperplasia. The reason for the inconsistency in our data and previous studies from Sudan is likely due to the characteristics of the study population and the delayed onset of symptoms. Previous studies were conducted at a tertiary care facility specializing in pediatric endocrinology.

Infants with CAH tend to experience symptoms like diarrhea, vomiting (salt loss), high blood pressure due to salt retention, and are promptly diagnosed and treated with cortisone therapy. Such cases are commonly referred to pediatric endocrinology, which could explain the difference in results between our study and previous ones.

There was a high percentage of consanguineous marriage in our study population, as 85.0% of the parents were consanguineous. This eventually led to an increase in the number of 5αR2D transmitted in the autosomal recessive mode of inheritance. It is worth noting that five siblings from 2 families also showed familial 5αR2D, again indicating the impact of consanguineous marriage on the profile of DSD among Sudanese patients. All five patients (2 siblings and three first-degree cousins) presented complaining of ambiguous genitalia. Similar previous reports described four ambiguous genitalia patients from the same family, and 46,XY DSD in two familial cases[16,17]. It would have been exciting to explore further the molecular profile of the twining family of two families to shed more light on the phenotype variations seen in siblings with 5αR2D. Unfortunately, the molecular sequencing of the two families with familial 5αR2D was omitted due to the high cost and shortage of funding in Sudan.

In 38.3% of the patients, the results showed a clash between the sex of rearing (grew as female) and the genotype (XY). In contrast, phenotype-genotype compatibility was seen in only 18.4% of adult patients. This paradox may be because sex assignment was irrationally determined at birth by an unqualified labor attendee or even by an inexperienced medical practitioner who failed to recognize and refer the patient for proper diagnosis and management[18]. The afterbirth wrong assignment, especially for the XY DSD group, has a severe consequence, especially in Sudan, where female genital mutilation risk is very high for those patients during childhood. Although female genital mutilation (FGM) has been outlawed since 1946 in Sudan, this brutal act is still widely practiced, with incidence reaching 89% in northern Sudan, and 86.6% of women aged 15–49 have undergone some form of FGM[19]. The present study revealed that 20 patients (33.3%) had 46 XY karyotypes grown as females. Of those, 15 patients (75%) were subjected to female genital mutilation. Circumcision usually alters the genitalia anatomy in the cases of 46 XY DSD patients misdiagnosed as females by non-experienced practitioners, where male external genitalia are usually removed, making it difficult to diagnose and manage those patients, and hence, the damage remains irreversible. A previous study from Sudan reported five cases of 46 XY DSD who were wrongly assigned as females; all the patients were subjected to female genital mutilation resulting in complete or partial amputation of the penis despite strong male orientation after the Minnesota Multiphasic Personality Inventory (MMPI)[14]. Another study also reported a Sudanese patient of XY DSD who grew up as a female and was subjected to Type IV FGM (Pharaonic circumcision)[20].

Molecular analysis of the SRY gene was positive in 60.0% of the patients and negative in the remaining 40%, showing complete accordance with the final diagnosis and supporting the notion that the test is appropriate to aid in detecting the presence or absence of the SRY gene in XX males and XY females.

The SRY gene test analysis in this study helped determine the DSD type, especially among children. It has the advantage of quick detection, lower cost than cytogenetics, and more sensitivity in children than ultrasound[21]. The results of the present study confirm that the use of SRY analysis and G-banding analysis is cost-effective and helpful for the primary diagnosis of DSD among children.

Most Sudanese tribes originated from three main linguistic groups: The Nilo-Saharan, the Afro-Asiatic, and Niger-Kordofanian. According to the present study, 70.0% of the patient were from the Afro-Asiatic group, 28.3 % from the Nilo-Saharan, and 1.7 % were from the Niger Kordofanian group. Unfortunately, at the moment, we cannot provide an explanatory answer for this observation, and perhaps more studies on the role of autosomal genes in the pathogenesis of the DSD provide answers.

DSD in developing countries represents a range of complexities and necessitates a comprehensive medical team, including a pediatric endocrinologist, pediatric surgeon, gynecologist, and physiatrist[22]. Accurate diagnosis relies on access to advanced laboratory facilities capable of conducting hormonal profiles, cytogenetics and molecular analysis, and top-notch imaging facilities. Unfortunately, many African regions with limited health resources lack these essential facilities[23].

Compounding the issue are social stigmas surrounding the disease, the high cost of investigations, the high percentage of Consanguineous marriage, which leads to an increase in cases, the high number of unnoted cases at birth leading to late presentation, and finally, the significant risk of female genital mutation for those wrongly assigned as female at birth.

## Conclusion

As per the present study's findings, disorders of sex development (DSD) have a distinct profile marked by late presentation, with the majority of patients reporting to medical facilities after reaching puberty. Interestingly, many of these patients come from the linguistic Afro-Asiatic tribes. The study also found that 5αR2D-DSD is the most frequent than other types of DSD, potentially due to increased consanguinity among parents and late diagnosis of patients. Overall, the study provides crucial insights into the characteristics of DSD patients presented late and underscores the importance of prompt diagnosis and intervention.

## Competing interest

None

## Ethical consideration

The Ethics Committee at Al Neelain University has granted ethical approval for the study. Patients or parents of underage patients have provided written consent to participate in the study. The study involved conducting necessary medical history, physical examination, hormonal profile, pelvic ultrasound, cytogenetic and molecular analyses. The clinical data and results were coded and anonymized to maintain confidentiality.

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## Data availability

All data are available upon request

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## Contribution of authors

Manal M. E. Awad Alkareem and Imad Fadl-Elmula conceived the project, while clinical and genetic data collection was carried out by Manal M. E. Awad Alkareem, Samia M. Ahmed, Rayan Khalid, and Houda M. Dguimi. Analysis and interpretation of the data were performed by Manal ME Awad Alkareem, Rayan Khalid, Huda, and Samia Mahdi. Manal M. E. Awad Alkareem, Houda M. Dguimi, and Samia M. Ahmed wrote the initial manuscript draft, with input and revisions from Rayan Khalid and Imad Fadl-Elmula. All authors have approved the final draft of the manuscript.

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