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Case Report

Unraveling of Diagnostic Odyssey in A Girl with Primary Amenorrhea: A case report

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Abstract

Primary amenorrhea may result from congenital abnormalities in the development of the gonads, genital tract, or external genitalia or from a disturbance within the hypothalamic-pituitary-ovarian axis. Gonadal dysgenesis is a disorder of sex development in which the diagnosis is based on the histology of gonads and is the main cause of primary amenorrhea. Optimal protocol of management for phenotypic female with 46, XY gonadal dysgenesis involves prophylactic gonadectomy at diagnosis.

Case Presentation: The patient was referred to our hospital at the age of 15 years old for primary amenorrhea. She was obese with no secondary sex signs. Gynecologic examination revealed a normal vagina and clitoris. Rectal Toucher examination revealed no internal genitalia structure. The laboratory data: FSH levels were above normal range, LH and testosterone levels were within normal range. In pelvic ultrasonography uterus and vaginal structure and testis were not visualized. Cytogenetic and *AR* gene analysis found a 46, XY karyotype and no pathogenic variants. On laparoscopy, Mullerian structure and Wolffian remnant structure were identified and biopsies were performed. Based on histopathological examination and immunohistochemical markers of the right and left gonad showed the impression of Malignant Mixed Germ Cell-Sex Cord Stromal Tumor. SRY gene examination was positive. Examination of other DSD gene analysis has not been done. A Second laparoscopy for gonadectomy and removal of Mullerian and Wolffian remnant structure were performed.

Conclusion: Chromosomal analysis should become the first line testing in primary amenorrhea followed by advanced molecular test. Multidisciplinary management is recommended for DSD cases.

Keywords: Primary amenorrhea, Gonadal dysgenesis, Disorders of sexual development, Genetic mutation
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INTRODUCTION

Primary amenorrhea is the failure of menses to occur by age 16 years, in the presence of normal growth and secondary sexual characteristics or failure of menses to occur by age 14 in the absence of normal growth and secondary sexual characteristic. Primary amenorrhea is most common caused by gonadal dysgenesis (approximately 30-40%), followed by Mullerian agenesis, and androgen insensitivity syndrome.[1]

The term disorders of sex development (DSD) are defined by "congenital conditions in which development of chromosomal, gonadal or anatomical sex is atypical."[2] DSD classification proposed by the multidisciplinary conference in Chicago 2005 are 46, XX DSD, 46, XY DSD and sex chromosomal DSD.[2-4]

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The overall incidence rate for DSD is 1 in 5.500, genetic deformities during sexual differentiation is the predominant caused.[3] DSD are commonly correlated with genital ambiguity, that often caused physical and psychological problems. Patients with XY pure gonadal dysgenesis, XY mixed gonadal dysgenesis and XY androgen insensitivity syndrome known to be DSD in XY phenotypic females.[5]

Presentation and distinctive measures of XY pure gonadal dysgenesis are presence of female external and internal genitalia with 46, XY karyotype. The patients usually have no Turner stigmata and may present with uterus and oviducts and nonappearance of follicles in the gonadal line. The failure of gonad differentiation into functional testes in 46, XY pure gonadal dysgenesis DSD caused the reproductive tract formed into a female pathway.[6] Mixed gonadal dysgenesis is identified in patients with a one sided testis (frequently intra-abdominal), a contra-lateral gonadal line, formation of pertinacious Mullerian duct with differing levels of undermasculinization.[5] The great number of patients diagnosed with gonadal dysgenesis have an evident anomaly implicating X chromosome, although mosaicism also have been reported with Y chromosomes.[1] Androgen insensitivity syndrome (AIS) is a rare X-linked disorders due to pathogenic variants in the androgen receptor (AR) gene (Xq11–q12).[7,8] Molecular diagnosis was made by identification of pathogenic variants in the *AR* gene.[9]

The scarcity of 46 XY GD patients with an incidence rate 1 in 100.000 in which most cases caused by genetic disorders.[10] Patients with GD conventionally have mutations on the testis-determining gene SRY[11], and other genes such as *WT1, DHH, NR5A1, SOX9, GATA4, FOG2/ZFPM2*, and *MAP3K1*, as well as chromosomal adjustment including deletions of chromosome 9p and duplications of Xp22.[12,13] After the diagnosis is established, immediate gonadectomy must be performed, because there is 30% risk of malignant gonadal line alterations in patients with 46, XY complete gonadal dysgenesis (CGD) at the age 40 years. Gonadoblastoma is the most common gonadal tumor in 46, XY CGD. Most dysgerminoma are gonadoblastoma associated with malignant germ cell tumors with an incidence of 50%-60% of cases.[14] It is very important to be able to carry out a careful initial assesment and early identification, because there is a risk for malignancy alterations that can occur in early life and increases sharply with age even though the history of the origin of gonadoblastoma is unclear.[15] Gonadal histology is the basis for establishing a diagnosis of gonadal dysgenesis which is a disorder of sex development. Preventive gonadectomy at diagnosis is the most ideal management protocols for phenotypic females with 46, XY GD. In this case report we present a case of primary amenorrhea with gonadal dysgenesis

CASE REPORTS

The patient was referred at the age of 15 years old for primary amenorrhea. She was obese (Asian criteria), weighing 77,7 kg for a height 166 cm (BMI 28 kg/m^2). Her pubertal status based on Tanner scale, was A1, M1, P1. Gynecologic examination revealed a normal vagina and clitoris. Rectal Toucher examination revealed no internal genitalia structure was found. The laboratory data: FSH levels 47.15 mIU/ml (normal range: 0.57-8.77 mIU/ml), LH levels 10.53 mIU/ml (normal range: ≤ 15.97 mIU/ml), testosterone level 17.15ng/dl (normal range: 14.12-48,99 ng/dl). In pelvic ultrasonography uterus and vagina structure were not visualized and no testes structure were identified. Approval from the medical ethics committee of Faculty of Medicine Diponegoro University and informed consent from the patients and her parents prior to their participation in this case report were obtained. Her karyotype was 46, XY. *AR* gene analysis was no mutation. On laparoscopy, Mullerian structure (uterus, fallopian tubes, and gonads) was identified (fig. 1), a vas deferens-like structure in the lateral pelvic wall was found and biopsies were performed. Based on histopathological examination and immunohistochemical profile of the right and left gonad showed seminiferous tubules containing Leydig cells and no visible epithelial layer of the ductal deferens with the impression of Malignant Mixed Germ Cell-Sex Cord Stromal Tumor. Mullerian structures were not confirmed. Immunohistochemical markers for gonadal germ cell tumors are positive for PLAP and CD117. Further analysis of DSD genes, SRY gene alleles was found. Examination of other DSD gene analysis has not been done. The patient was consulted to the urology department for a second laparoscopy, gonadectomy and removal of Mullerian/Wolffian remnant structure were performed. Gonadal dysgenesis with gonadoblastoma and Malignant Germ Cell Tumor was confirmed.

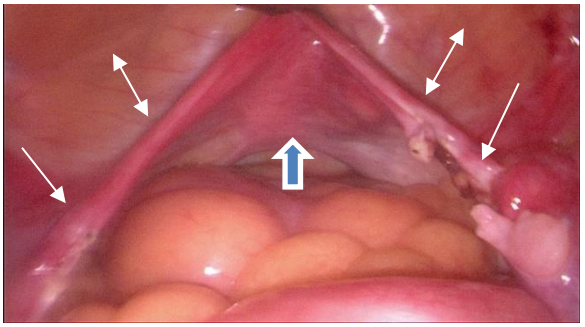

Figure 1. Laparoscopic of Mullerian structure.
Left/Right streak gonad(arrow), left/right fallopian tubes (double arrow), uterus (up arrow).

DISCUSSION

Performing cytogenetic investigation procedures is a compelling reason for every patient experiencing hypergonadotropic hypogonadism related with external and internal genitalia in female even when there is no clinical characteristic of Turner syndrome, but it is difficult to gain karyotypes. This was done considering the significant proportion of XY patients among those with GD. Current research shows there is a high incidence of XY patients among those with GD where more than one third of the specimen has a chromosome structure, which shows the prominence of analyzing the karyotype of patients with this clinical feature.[16]

The phenotype of female with a blind vaginal pouch and no uterus is a distinctive sign of Complete Androgen Insensitivity Syndrome (CAIS). The existence of testes with histology according to testicular differentiation, 46, XY karyotype, and nonappearance of Müllerian duct

remnants are needed to establish the diagnosis of AIS.[17] Primary amenorrhea is highly prevalent complaint that requires medical support during puberty. The clitoris and labia minora are underdeveloped in some cases.[9] The proband referred to our clinic by reason of primary amenorrhea. Based on the clinical evaluation supported by hormonal assay and cytogenetic analysis, the patients were clinically diagnosis had CAIS and the *AR* gene analysis were performed. The analysis was only covering 8 exons of *AR* gene, to determine whether mutations were in the intron or regulatory region, it should proceed to a more advanced examination using the whole gene sequencing. Jaaskelainen (2012) declared that there are a large number of diverse mutations in the *AR* gene related with AIS. Missense mutations are the majority of mutations and most affect the ligand-binding domain of the *AR*. Exon 1 novel mutations values will increase if this exon is orderly filtered for mutations. Several cases shown where there were AIS characteristics in vivo and in vitro but no mutations in the *AR* coding area can be found. There has never been a case with mutations that occurred in the *AR* promoter region even though this could happen theoretically.[17] Listyasari, et al (2019) reported clinical, imaging, hormonal, and molecular analysis among girls with primary amenorrhea, confirmed have CAIS. [18] The clinical findings of this patient with no secondary sex signs, and from the molecular analysis of this patient, however, revealed no pathogenic variants in all exon, and diagnostic laparoscopy identified Müllerian structure, consequently the diagnosis of CAIS is excluded.

In pelvic ultrasonography of the patient showed that uterus and vaginal structure were not visualised and no testis structure were identified. The facts provided by pelvic MRI are superior to ultrasonography in recognizing Mullerian structures.[19]

The majority types of tumors found in patients with DSD are gonadoblastoma and dysgerminoma.[20] It is a well-established association between the Y-derived chromosome materials and the development of gonadoblastoma. It is known approximately in 15 – 20% cases of X/XY patients and 30% cases of patients with XY gonadal dysgenesis will develop gonadoblastoma.[21] In this case reports, the patient was confirmed for gonadoblastoma and Malignant Germ Cell Tumor. Gonadoblastoma described as an in-situ germ cell neoplasm that has potential for developing malignant germ cell tumors such as dysgerminoma.[22,23] Gonadoblastoma can be identified in gonadal tissue with undifferentiated or immature testis differentiation.[4] It was hypothesized from scientist that several factors involved in the evolution of gonadoblastoma during the gonad differentiation process were identical to factors involved in the growth of dysgenetic gonads.[24] At present there are no genes that act as oncogene and are involved in the development of the gonadoblastoma locus on the Y chromosome. Patients with sex reversal showing pathogenic variants in *WT1* and *SOX9* genes, 9p deletions can have this tumor.[25,26]

Comprehension of DSD category, age, and level of individual pubertal status is needed to make an appropriate timing of gonadectomy that will become a prominent point in DSD managements. Individuals with 46, XY GD and germ cell tumors have an incidence rate approximately at 25-33%.[27,28] There was a strong recommendation to do an early gonadectomy at diagnosis.[5] In this case, patient was 15 years old and based on the histopathological and immunohistochemical findings from the first laparoscopic biopsies showed malignancy. Evidence of gonadal germ cell tumors is confirmed by immunohistochemical marker staining using PLAP and CD117.[29] Therefore we performed a second laparoscopy for gonadectomy and removal of the Mullerian and Wolffian structure. Gonadal dysgenesis with gonadoblastoma and Malignant Germ Cell Tumor was confirmed based on anatomic pathology result.

Patients and their families frequently feel pressure after the establishment of the DSD diagnosis. Management collaboration between psychological care and counselling by psychologist is needed other than appropriate medical and surgical care.[30] As to our DSD patient, a comprehensive multidisciplinary team is essential in order to provide the best of care for patient with DSD followed by genetic counseling and psychological support for the patient, and her family members. In this patient, gender assignment as female was made based on evaluation of gender identity and gender role by the multidisciplinary team in our center, taking into account the patient's wish.[31]

In collaboration with the urology department, patient's improvement was assessed for 3 months and planned to be given substitution of estrogen to promote her secondary sex development, to reduce risk of osteoporosis and cardiovascular disease. Furthermore, for long-term treatment, psychological care will be carried out.

CONCLUSION

Chromosomal analysis should become the first line testing in primary amenorrhea. A comprehensive and multidisciplinary approach of DSD is paramount for clinical managements for primary amenorrhea patients. Genetic etiology such as mutation involving the testis-determining gene SRY and other genes involved in sex determination should be elucidated through advanced molecular analysis such as gene panel using massive parallel sequencing.

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REFERENCES

1. Fritz MA, Speroff L. Amenorrea. Clinical gynecologic endocrinology and infertility. 9th ed. Wolters kluwers.2020;10.
2. Lee PA, Houk C, Ahmed SF, Hughes I. Consensus statement on management of intersex disorders. International consensus conference on intersex.2006. 118(2).

3. Hughes I, Houk C, Ahmed SF, Lee PA, LWPES Consensus Group, ESPE Consensus Group.Consensus statement on management of intersex disorders. Arch Dis Child 2006; 91:554-563.
4. Hersmus R, Stoop H, White SJ, Drop SL, Oosterhuis JW, Incrocci L,et al . Delayed recognition of disorders of sex development (DSD): a missed opportunity for early diagnosis of malignant germ cell tumors. Int J Endocrinol.2012; 671209.
5. Liu AA, Shi HY, Cai ZJ, Liu A, Zhang D, Huang HF,et al . Increased risk of gonadal malignancy and prophylactic gonadectomy: a study of 102 phenotypoc female patients with Y chromosome or Y-derived sequences. Human Reprod. 2014: 29:7.
6. Takai Y, Tsutsumi O, Harada I, Morita Y, Momoeda M, Fukushima Y, Taketani Y. A case of XY pure gonadal dysgenesis with 46, XYp-/47, XXYp-karyotype whose gonadoblastoma was removed laparoscopically. Gynecol Obstet Invest 2000; 50:166–169.
7. Lubhan DB, Joseph DR, Sullivan PM, Willard HF, French FS, Wilson EM. Cloning of the human androgen receptor complementary DNA and localization to the X chromosome. Science.1988; 240:327–330.
8. Radpour R, Falah M, Aslani A, Zhong XY, Saleki A. Identification of a critical novel mutation in the exon 1 of androgen receptor gene in 2 brothers with complete androgen insensitivity syndrome. J Androl 2009; 30:230 – 232.
9. Mendoza N, Motos MA. Androgen insensitivity syndrome. Gynecol Endocrinol 2013; 29:1 -5.
10. Bastian C, Muller JB, Lortat-Jacob S, Nihoul-Fekete C, Bignon-Topalovic J, McElravey K, et al. Genetic mutation and somatic anomalies in association with 46, XY gonadal dysgenesis. Fertil Steril.2015; 103:5.
11. McElraevy K, Vilain E, Abbas N, Costa JM, Souleyreau N, Kucheria K, et al. XY sex reversal associated with a deletion 5' to the SRY HMG box in the testis-determining region. Proc Natl Acad Sci U S A. 1992; 89:11016-20.
12. Bashamboo A. McElreavey K. Gene mutations associated with anomalies of human gonad formation. Sex Dev 2013; 7: 126-46.
13. White S, Ohnesorg T, Notini A, Roeszler K, Hewitt J, Daggag H, et al. Copy number variation in patients with disorders of sex development due to 46, XY gonadal dysgenesis. Plos One 2011; 6: e 177793.
14. Doherty LF, Rackow BW. Abnormal streak gonads in 46, XY complete gonadal dysgenesis. Fertil Steril. 2011; 96:6.
15. Manuel M, katamya PK, Jones HW Jr. The age of occurance of gonadal tumours ini intersex patients with a Y chromosome. Am J Obstet Gynecol 1976; 124: 293-300.
16. Rocha VBC, Guerra-Junior G, Marques-de-Faria A, De Mello MP, Maciel-Guerra AT. Complete gonadal dysgenesis in clinical practice: the 46, XY karyotype account for more than one third of cases. Fertil Steril. 2011; 96:6.
17. Jaaskelainen J. Molecular biology of androgen insensitivity. Elsevier. 2011.
18. Listyasari NA, Robevska G, Santosa A, Bouty A, Juniarto AZ, Van den Bergen J, Ayers KL, et al. Genetic Analysis Reveals Complete Androgen Insensitivity Syndrome in Female Children Surgically Treated for Inguinal Hernia. J Invest Surg. 2019; 0: 1-7.
19. Ahmed SF, Achermann JC, Arlt W, Balen AH, Conway G, Edwards ZL, et al. UK guidance on the initial evaluation of an infant or an adolescent with a suspected disorder of sexual development. Clin Endocrinol (oxf). 2011; 75: 12-26.
20. Gravholt CH, Fedder J, Naeraa RW, Mu¨ ller J. Occurrence of gonadoblastoma in females with Turner syndrome and Y chromosome material: a population study. J Clin Endocrinol Metab 2000; 85: 3199 – 3202.
21. Verp Ms, Simpson JL. Abnormal sexual differentiation and neoplasia. Cancer Genet Cytogenet. 1987; 25: 191-218
22. Scully RE. Gonadoblastoma. A review of 74 cases. Cancer 1970; 25:1340 – 1356.
23. Jørgensen N, Muller J, Jaubert F, Clausen OP, Skakkebaek NE. Heterogeneity of gonadoblastoma germ cells: similarities with immature germ cells, spermatogonia and testicular carcinoma in situ cells. Histopathology 1997; 30:177 – 186.
24. Mazzanti L, Cicognani A, Baldazzi L, Bergamaschi R, Scarano E, Strocchi S, Nicoletti A, Mencarelli F, Pittalis M, Forabosco A et al. Gonadoblastoma in Turner syndrome and Y-chromosome-derived material. Am J Med Genet 2005; 135A: 150–154.
25. Lahn BT, Page DC. Functional coherence of the human Y chromosome. Science 1997; 278:675–680.
26. Livadas S, Mavrou A, Sofocleous C, van Vliet-Constantinidou C, Dracopoulou M, Dacou-Voutetakis C. Gonadoblastoma in a patient with del (9) (p22) and sex reversal: report of a case and review of the literature. Cancer Genet Cytogenet 2003; 143: 174–177.
27. Troche V, Hernandez E. Neoplasia arising in dysgenetic gonads. Obstet Gynecol Surv 1988; 41: 74–79.
28. Cools M, Drop SL, Wolffenbuttel KP, Oosterhuis JW, Looijenga LH. Germ cell tumors in the intersex gonad: old paths, new directions, moving frontiers. Endocr Rev 2006; 27: 468 – 484.
29. Gao Y, Jiang J, Liu Q. Extragonadal malignant germ cell tumor: a clinicopathological and immunohistochemical analysis of 48 cases at a single chonese institution. Int J Clin Exp Pathol. 2015; 8(5): 5650-5657.
30. Hughes I, Deeb A. Androgen resistance. Best Pract Res Clin Endocrinol Metab 2006; 20: 577 – 598.
31. Listyasari NA, Santosa A, Juniarto AZ, Faradz SMH. Multidiscipinary Management of Disorders of Sex Development in Indonesia, A Prototype for Develpoing Country. J Biomed Trans Res. 2017; 01: 17-22