Investigation and treatment of primary amenorrhoea

Tim Child

Abstract
Primary amenorrhoea is most commonly caused by constitutional delay or genetic or structural factors. However, any cause of secondary amenorrhoea can also present before menarche and lead to primary amenorrhoea. This review addresses the path of normal puberty and the causes and appropriate investigations and treatments of primary amenorrhoea in a systematic manner. The importance of considering the oestrogen state of the woman is stressed to ensure that, where necessary, replacement therapy is given to maximize bone mass and reduce the lifetime risk of fractures.

Keywords androgen insensitivity syndrome; constitutional delay; gonadal agenesis; mullerian agenesis; primary amenorrhoea; puberty; Turner syndrome

Introduction
There are a multitude of causes of absence of menstruation (or amenorrhoea). The division into primary or secondary amenorrhoea is somewhat arbitrary since any cause of secondary amenorrhoea can cause primary amenorrhoea (such as pregnancy). Primary amenorrhoea can be defined as amenorrhoea in a patient otherwise expected to have regular periods. Secondary amenorrhoea is amenorrhoea in a woman who has already established menstruation though now absent for 3–6 months. Pathology presenting before menarche will present as primary amenorrhoea and after menarche as secondary amenorrhoea. However, chromosomal or structural causes more commonly underlie primary than secondary amenorrhoea. This article will describe the causes, investigation and treatment of primary amenorrhoea. An initial short review of normal puberty will assist in understanding relevant pathology and the appropriate investigations and treatment.

Normal female puberty
North American studies from the 1980s established the average age of pubertal onset, as indicated by breast budding, as 10.7 years (SD 1 year) and average age of menarche as 12.7 years (SD 1.3 years). Recent studies suggest a trend towards earlier puberty possibly due to higher BMI scores.

Puberty begins with the pulsatile release of GnRH from neurons within the hypothalamus, with the subsequent secretion of LH and FSH. Factors involved in the initial ‘awakening’ of pulsatile GnRH release include the neuropeptide kisspeptin acting on the G protein-coupled receptor 54 (GPR54) and the action of leptin. Leptin, a protein produced by the adipocyte, acts on the hypothalamus to regulate food intake, energy expenditure and body weight. Leptin may act as a metabolic gate, allowing increased gonadotropin secretion when leptin levels are sufficient. About one year prior to breast budding the amplitude of LH peaks increase during sleep. LH leads to ovarian production of androstenedione and FSH to its ovarian conversion to oestradiol. The increasing oestradiol levels lead to enlarging breast tissue and, through its influence on bone growth and epiphyseal fusion, the pubertal growth spurt.

Menarche usually occurs during Tanner stage 4 of breast development though it can take 5–7 years for the endocrine axis to mature and for the establishment of regular menstrual cycles. In the first year after menarche 50% of cycles are anovulatory though 80% fall in the range of 21–45 days duration.

Causes of amenorrhoea
Table 1 lists the causes of primary amenorrhoea organized by the presence or absence of breast development and high or low levels of serum FSH. The presence of breast tissue signifies previous (and possibly present) oestrogen action. It is clear that, since primary amenorrhoea is itself not common, many of the causes listed will be rarely seen in a normal reproductive medicine clinic. The numerous other pathologies described as causes of primary amenorrhoea will therefore be extremely uncommon. Pregnancy must be excluded and the possibility of constitutional (non-pathological) delay always considered.

Table 1 Common causes of primary amenorrhoea (adapted from ASRM Practice Committee. Amenorrhoea. Fertil Steril 2008)

<table>
<thead>
<tr>
<th>Cause</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>No breast development and high FSH</em></td>
<td></td>
</tr>
<tr>
<td>46 XX</td>
<td>15%</td>
</tr>
<tr>
<td>46 XY</td>
<td>5%</td>
</tr>
<tr>
<td>Abnormal karyotype</td>
<td>20%</td>
</tr>
<tr>
<td><em>No breast development and low FSH</em></td>
<td>~30%</td>
</tr>
<tr>
<td>Constitutional delay</td>
<td>10%</td>
</tr>
<tr>
<td>Prolactinomas</td>
<td>5%</td>
</tr>
<tr>
<td>Kallman syndrome</td>
<td>2%</td>
</tr>
<tr>
<td>Other CNS</td>
<td>3%</td>
</tr>
<tr>
<td>Stress, exercise, weight loss, anorexia</td>
<td>3%</td>
</tr>
<tr>
<td>CAH</td>
<td>3%</td>
</tr>
<tr>
<td>Other</td>
<td>1%</td>
</tr>
<tr>
<td><em>Breast development</em></td>
<td>~30%</td>
</tr>
<tr>
<td><em>Uterus absent</em></td>
<td></td>
</tr>
<tr>
<td>Mullerian agenesis</td>
<td>10%</td>
</tr>
<tr>
<td>Androgen insensitivity</td>
<td>9%</td>
</tr>
<tr>
<td><em>Uterus present</em></td>
<td></td>
</tr>
<tr>
<td>Vaginal septum</td>
<td>2%</td>
</tr>
<tr>
<td>Imperforate hymen</td>
<td>1%</td>
</tr>
<tr>
<td>PCOS</td>
<td>3%</td>
</tr>
<tr>
<td>Constitutional delay</td>
<td>8%</td>
</tr>
</tbody>
</table>

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When and how to investigate

Recent American Society for Reproductive Medicine guidelines suggest evaluation of primary amenorrhoea under the following circumstances:

- Failure to menstruate by age 15 in the presence of normal secondary sexual development (2 SD’s above the mean age)
- Within 5 years after breast development if that occurs before age 10
- Failure to initiate breast development by age 13 (2 SD’s above the mean age)

Traditionally age 16 was used as the trigger point for evaluation.

Consultation and examination

The approach taken to the consultation needs to recognize the sensitivities of adolescents, particularly those with medical problems. If she perceives the consultation as difficult or traumatic this may have long lasting ill-effects. The patient is usually seen with a parent and plenty of consultation time allowed to build up rapport and allow a full and unhurried discussion. The management of patients with primary amenorrhoea should occur in a specialized clinic that can offer a multidisciplinary approach to care.

A full patient history is taken with particular focus on growth and development and any signs of puberty. Often only a limited examination need be undertaken initially, in the presence of a chaperone, since the results of further investigations may dictate subsequent actions. Box 1 lists the main points. The genital examination is abnormal in around 15% of patients with primary amenorrhoea. If necessary examination under anaesthetic may be indicated to help make the diagnosis.

Investigations

Pregnancy should always be excluded with a urine hCG test. Imaging of the pelvis with a transabdominal (in non-sexually active) and/or transvaginal ultrasound scan will confirm the presence or absence of a uterus and normal sized ovaries. Alternatively CT or MRI can be used.

Serum estimation of FSH, prolactin and TSH will exclude ovarian or hypothalamic failure, pituitary adenomas and thyroid disease.

Box 1

Examination for primary amenorrhoea

**General**

- Weight, height, body mass index
- Blood pressure
- Clinical thyroid status
- Dysmorphic signs
- Tanner breast and axillary hair stages

**Abdomen and pelvis**

- Mass arriving from pelvis
- Groin nodes/herniae

**Perineum**

- Often only inspection, particularly if not sexually active
- Pubic hair distribution, clitoral size, hymen, evidence of oestrogenization

If the serum FSH is raised (>20 IU/L) or the uterus absent then a serum karyotype is indicated. With a raised FSH the main diagnoses are premature ovarian failure (46, XX) or Turner syndrome (46, XO). If the uterus is absent then the likely diagnoses are androgen insensitivity syndrome (46, XY) or mullerian agenesis (46, XX).

An algorithm for investigation is presented in Figure 1.

Investigation results and treatment

**Absence of secondary sexual characteristics**

In the absence of secondary sexual characteristics including breast development and pubic hair the serum FSH level will be indicative of the underlying cause. Normal FSH levels are in the range of around 3–10 IU/L. Levels below this suggest hypogonadotropic hypogonadism (no ‘drive’ from the hypothalamus/pituitary to the ovaries) and levels above 20 IU/L suggest hypergonadotropic hypogonadism (absence of functioning ovarian tissue).

**Hypogonadotropic hypogonadism**: whilst there are a number of possible causes of hypogonadotropic hypogonadism, constitutional delay is the most common factor. It should be noted that constitutional delay may be difficult to differentiate from ‘pathological’ causes of hypogonadotropic hypogonadism. Constitutional delay may be associated with a positive family history, short stature, delayed secondary sexual characteristics and delayed epiphyseal maturation (identified by X-ray aging of the hand). The final height prognosis remains in the appropriate range for the parental centiles (easily calculated from growth charts).

Prolactinomas are easily excluded through measurement of serum prolactin levels and, if levels are raised >1500 mU/L, through MRI or CT imaging of the head. Hyperprolactinaemia causes hypogonadism through inhibition of GnRH release. If a micro or macro adenoma is diagnosed then endocrine review is indicated for subsequent management, usually involving dopamine-agonist treatment. Iatrogenic causes of hyperprolactinaemia include medications such as some anti-psychotics, domperidone and metoclopramide.

Kallman syndrome is rare (1:40 000—50 000 females) and associated with defects in olfactory bulb development. Genetic inheritance can be X-linked (KAL1), autosomal dominant (KAL2) or autosomal recessive (KAL3). Kallman syndrome is significantly more common in males (1:8000). Women with Kallman syndrome may have anosmia as well as amenorrhoea and low gonadotropins due to GnRH deficiency.

Other causes of hypogonadotropic hypogonadism include cranial tumours such as craniopharyngiomas, gliomas, germinomas, hamartomas and teratomas. They may cause additional symptoms such as headaches or other neurological sequelae and clearly require neurosurgical attention. Treatment for cranial tumours in childhood, including neurosurgery or cranial radiotherapy, may irreversibly damage hypothalamic-pituitary function and lead to primary amenorrhoea.

Weight loss and anorexia are again uncommon but important causes of primary amenorrhoea. The prevalence of anorexia nervosa in young women is 0.3–0.5% with the highest incidence in adolescent girls between 15 and 19 years. As described previously, there exists a link between weight and amenorrhoea. However, theories suggesting a critical percentage of body fat required for onset and maintenance of menses have been challenged. Studies
clearly demonstrate that some women with low levels of body fat have regular menses and, in both athletes and women with anorexia nervosa, studies do not always confirm a difference in percentage of body fat between those who menstruate and those who don’t. Furthermore, around 20% of women with anorexia develop amenorrhea before significant weight loss. It has been suggested that this is mediated by leptin’s response to calorific restriction and a negative energy balance prior to significant weight loss.

A diagnosis of anorexia clearly requires psychological and psychiatric input and management. Low body weight may also be due to high levels of exercise, which alone may also lead to reduced hypothalamic drive. More than 90% of adolescents and young adults with anorexia nervosa have reduction of bone mineral density at one or more points. The long term fracture risk is increased 2–3-fold compared to controls. Exercise-induced amenorrhea is similarly associated with reduction in bone mass and increased fracture risk.

Chronic debilitating illness including malabsorption syndromes and renal or liver disease can lead to hypogonadotropic hypogonadism. Chronic illnesses may affect nutritional, behavioural, hormonal and metabolic aspects and so cause amenorrhea through a number of pathways. In many chronic illness states there may be a discrepancy in calorific intake compared to requirements, and the mechanism of amenorrhea can be similar to that found in anorexia and excessive exercise states. Congenital adrenal hyperplasia occurs in classic and non-classic forms, the latter can present in early adolescence with amenorrhea.

In the absence of any of the above likely diagnoses then constitutional delay is most likely. A thorough family history may also point to this diagnosis if female relatives have had a similar delay in menstruation. Twin studies reveal that genetic factors are the largest single factor accounting for variability in the timing and tempo of puberty. If considered likely then ‘watchful waiting’ and reassurance is an option, or puberty can be induced.

Treatment: ideally treatment is aimed at the underlying identified cause. Successful treatment of cranial tumours (including pituitary adenomas) may lead to resumption of pubertal development.
Where relevant, an increase in weight or reduction in exercise may also lead to an improvement. If the hypogonadism is permanent (for instance after cranial surgery or radiotherapy) and puberty has not yet started then puberty needs to be induced using increasing doses of ethinyl-oestradiol under the care of a paediatric endocrinologist. A suitable regime may be oral ethinyl-oestradiol 2 mcg daily, increasing by 5 mcg every 6 months to 20 mcg, then conversion to a combined oral contraceptive pill. It is important to optimize skeletal growth and uterine growth, for possible later pregnancy, by inducing puberty at the appropriate time and with the appropriate drug regimen. Puberty can also be induced when constitutional delay is thought to be the likely diagnosis.

If hypogonadotropic hypogonadism persists then maintenance of sufficient oestradiol levels for optimal bone density, cardiovascular health, and to prevent numerous menopausal symptoms and complications can be achieved through use of a combined oral contraceptive pill or a combined HRT preparation. This can be continued until fertility is desired. At that point ovulation is induced using daily sub-cutaneous injections of gonadotropins for around 2 weeks followed by timed sexual intercourse. Cumulative pregnancy rates are high, in the region of 70%.

**Hypergonadotropic hypogonadism**

The absence of secondary sexual characteristics in the presence of high serum levels of FSH suggests either the occurrence of ovarian failure prior to the onset of puberty or failure of development of normal ovarian tissue (gonadal dysgenesis). A serum karyotype is required, the most common abnormal finding being Turner syndrome (45, XO).

**Gonadal dysgenesis:** gonadal dysgenesis includes conditions where gonadal development is abnormal leading to streak ovaries, low oestradiol levels and high FSH levels. Turner syndrome (45, XO) is most commonly diagnosed though Swyer syndrome (46, XY) and pure gonadal dysgenesis (46, XX) are occasionally seen. Turner syndrome occurs in approximately 1:3000 female live births. In around half of cases there is complete or partial absence of an X chromosome (45, XO) but other findings include mosaicism (mixure of 45, XO and 46, XX cell lines). Clinical features of Turner syndrome include primary amenorrhoea and delayed puberty plus webbed neck and short stature, widely spaced nipples and low hairline, cubitus valgus (wide carrying angle) and renal and cardiac abnormalities (coarctation of the aorta).

**Premature ovarian failure:** there are numerous causes of premature ovarian failure, which can occur at any age, and if occur before puberty then present as primary amenorrhoea. Very often the cause is idiopathic but known causes include ovarian injury due to chemotherapy or radiation, or mumps oophoritis. Less common causes include galactosaeemia, female fragile X carriers, Trisomy 21 and autoimmune oophoritis. If an autoimmune cause is considered likely then thought should be given to testing and monitoring for other life threatening autoimmune conditions including Addison's disease, diabetes, hypothyroidism and hypoparathyroidism.

**Treatment:** any evidence of Y chromosome material requires surgical excision of the gonads, usually possible laparoscopically, to remove any risk of malignant change (gonadoblastoma).

Puberty may need to be induced as described before. Long term oestrogen replacement therapy is required with either combined HRT or the combined oral contraceptive pill. Pregnancy through oocyte donation as part of IVF treatment is possible for women with hypergonadotropic hypogonadism. There are additional risks of pregnancy for women with Turner syndrome due to the association with cardiac anomalies and careful pre- and ante-natal assessment and monitoring is required in a multidisciplinary environment.

**Secondary sexual characteristics present**

The presence of secondary sexual characteristics implies the presence of functioning gonads and circulating oestrogen. Pelvic imaging, performed most easily with ultrasound, will demonstrate the absence or presence of a normal uterus. Alternatively MRI or CT scanning can be employed. Most commonly, in this group of patients, the uterus will either be absent or very small and abnormal in which case the diagnosis is based on the karyotype results; Mullerian agenesis (46, XX) or androgen insensitivity syndrome (46, XY). If the uterus is present then either outflow obstruction, or other causes more usually associated with secondary amenorrhoea such as PCOS, are likely. Constitutional delay is also a possibility.

**Uterus absent or abnormal:** if the uterus is absent or very small and abnormal then the two main possible diagnoses are Mullerian agenesis (46, XX) or androgen insensitivity syndrome (46, XY).

Mullerian agenesis is the cause of approximately 10% of cases of primary amenorrhoea. It involves the congenital malformation of the genital tract resulting in absence of the upper part of the vagina, an abnormal or absent uterus and functioning ovaries with normal secondary sexual characteristics. Mullerian agenesis (often referred to as Rokitansky syndrome, or Mayer–Rokitansky–Kuster–Hauser syndrome in–full) occurs in 1:4000–5000 female births. It may result from the abnormal activation of either anti-mullerian hormone or its receptor through genetic mutation, causing regression of the mullerian duct.

Androgen insensitivity syndrome, due to X-linked recessive inheritance of a single gene, is very rare with an incidence of between 1:20 000 and 1:100 000. Serum testosterone levels are normal but there is usually a complete lack of binding at the androgen receptor sites. The testes are cryptorchid positioned within the inguinal canals or the abdominal cavity, without spermatogenesis, and produce anti-mullerian hormone leading to regression of the mullerian duct. Peripheral conversion of testosterone to oestrogen leads to normal breast development and there is generally minimal axillary and pubic hair with a short vagina.

**Uterus present:** if secondary sexual characteristics are present and a uterus identified on imaging then an outflow tract obstruction due to an imperforate hymen or transverse vaginal septum is possible and requires exclusion.

Imperforate hymen may be diagnosed in childhood or present later with primary amenorrhoea associated with cyclical abdominal pain. The vagina, and sometimes uterus, can become greatly distended with old blood. This may appear as a pregnant abdomen and can cause urinary retention. The hymen will be seen as bulging and bluish.

Complete transverse vaginal septum is due to incomplete fusion of the mullerian duct and uro-genital sinus portions of the
vagina. The majority occur in the upper vagina and the external genitalia appear normal. Other malformations of the rectum or uro-genital tract may co-exist.

Asherman’s syndrome, due to intra-uterine scarring following infection or uterine curettage is the commonest outflow obstruction cause of secondary amenorrhoea but would be extremely rare as a cause of primary amenorrhoea.

Once outflow obstruction has been excluded as a factor then causes more generally associated with secondary amenorrhoea need to be considered. Chief amongst these is polycystic ovary syndrome (PCOS).

Polycystic ovary syndrome is the commonest cause of secondary amenorrhoea and would be an unusual cause of primary amenorrhoea. PCOS is diagnosed when at least two of the following three factors are present: infrequent or absent menses, clinical or biochemical hyperandrogenism; ovaries of polycystic morphology on ultrasound scan. These criteria were developed for use in women rather than adolescents. The underlying pathophysiology of PCOS appears to be a degree of peripheral insulin resistance, which worsens with obesity. Hyperinsulinaemia disrupts ovarian folliculogenesis, increases production of ovarian androgens, and reduces hepatic production of sex hormone binding globulin. The end result is infrequent or absent periods and higher levels of circulating free androgens leading to acne and hirsutism. Not surprisingly, young adolescents with PCOS, who may be overweight and with skin problems, are recognized to report poorer quality of life scores than their peers. Weight loss of 5–10% is often sufficient to reverse the symptoms. Alternatively, use of a non-androgenic combined oral contraceptive pill may offer improved skin (through reducing free androgen levels by reducing ovarian androgen production and increasing sex hormone binding globulin) and regular withdrawal bleeds. Since PCOS is associated with normal levels of circulating oestrogen, its unopposed action on the endometrium can, after many years, lead to hyperplasia and possible malignancy.

**Treatment:** the diagnosis of Mullerian agenesis or androgen insensitivity syndrome can be devastating for a young woman and her family. Both diagnoses mean the impossibility of the woman carrying a child of her own. Androgen insensitivity syndrome also comes with the fact that the woman is genetically male (though phenotypically female). In previous years women were generally not given the karyotype result, perhaps only discovering the true diagnosis decades later. Such a consultation would be difficult for any patient but for a young adolescent then even more so. Clear and empathic explanations are required, in a multidisciplinary setting, with good psychological and counselling support.

At some point the tests should be surgically removed from patients with androgen insensitivity syndrome to prevent malignant change. This may be delayed until after the growth spurt has completed, and then oestrogen HRT commenced to be continued until the natural age of the menopause. Vaginal dilation using graded dilators for 6–12 months is very successful in achieving good functional vaginal length for women with androgen insensitivity syndrome or Mullerian agenesis. Surgery is not usually required for this purpose. Women with Mullerian agenesis generally have normally functioning ovaries and, following superovulation, oocytes can be retrieved, fertilized in-vitro, and the embryos transferred to the uterus of a surrogate. Unfortunately there are no fertility options open to women with androgen insensitivity syndrome.

If the uterus is present and outflow tract obstruction confirmed then surgery is required. A cruciate incision of an imperforate hymen will easily treat the problem. A transverse vaginal septum is much thicker and may be associated with rectal and urological abnormalities so surgery needs to be undertaken in a planned manner.

If constitutional delay is thought likely (see before) then puberty can be induced or ‘watchful waiting’ employed for a while longer.

**Oestrogen and bone mass**

In the adolescent with low levels of oestrogen (all those groups described above with absent breast development) the main medical risk is reduction in bone mineral density and increased risk of fracture. Peak bone mass in women occurs around the age of 25 years. Prolonged periods of low oestrogen levels prior to this may mean this peak density level is not reached, leading to a life-long increased risk of fractures. Puberty may therefore need to be induced and maintenance doses of oestrogen continued until fertility is an issue or the average age of the menopause reached.

**FURTHER READING**


**Practice points**

- When to investigate for primary amenorrhoea.
  - Failure to menstruate by age 15 in the presence of normal secondary sexual development.
  - Within 5 years after breast development if that occurs before age 10.
  - Failure to initiate breast development by age 13.

- Commonest causes in presence of breast development are constitutional delay, Mullerian agenesis, and androgen insensitivity syndrome.

- Commonest causes in absence of breast development are constitutional delay, Turner syndrome, ovarian failure, and prolactinomas.