Research Journal of Medical Sciences 7 (5-6): 134-147, 2013
ISSN: 1815-9346

The Endocrine Causes of Amenorrhoea-Abnormalities in the Hypothalamic-Pituitary-Ovarian Axis: A Review

Gabriela Camilleri
Our Lady of Mount Carmel Street, Munxar,
Gozo, MXR1311, Malta, Europe

Abstract: Menstruation depends on the presence of an intact uterus and vagina and on the presence of a functioning hypothalamic-pituitary-ovarian axis. Failure in any of these will invariably lead to a menstrual disorder like amenorrhoea or oligomenorrhoea. This review will focus on amenorrhoea, the absence of menses and the endocrine causes that could be responsible for it. In the first part of the review, an outline of the hormonal regulation of menstruation is given followed by a definition of primary and secondary amenorrhoea. Following this will be a detailed systematic evaluation of the conditions that lead to hormonal disturbances of the menstrual cycle, starting with conditions affecting the hypothalamus, pituitary and finally the ovaries. A brief outline of other endocrine disorders that can lead to amenorrhoea and menstrual cycle disturbances, like thyroid and adrenal disorders can also be found in this review. This study will conclude with an evaluation of the patient presenting with amenorrhoea the history, physical examination and laboratory tests that need to be done in specific clinical situations.

Key words: Menstruation, amenorrhoea, anorexia nervosa, hyperprolactinaemia, PCOS

INTRODUCTION

Hormonal regulation of the menstrual cycle: Menstruation is the cyclic, orderly sloughing of the uterine lining on behalf of the interactions of hormones produced by the hypothalamus, pituitary and ovaries (Popat *et al*., 2008). The latter three form an endocrine axis (known as the HPO axis) that functions via hormonal regulation and feedback loops.

The hypothalamus releases Gonadotropin-Releasing Hormone (GnRH) in pulses. This is transported to the anterior pituitary where it stimulates the gonadotrophs. Secretion of GnRH is regulated by a number of neurotransmitters including dopamine, endogenous opioids, norepinephrine, Gamma Amino Butyric Acid (GABA) and Corticotropin-Releasing Hormone (CRH). In turn, the gonadotrophs synthesize, store and secrete the gonadotropins Follicle-Stimulating Hormone (FSH) and Luteinising Hormone (LH) (Fig. 1) which in turn stimulate the ovaries to synthesize and secrete oestrogen, progestin and androgen.

In the follicular phase of the normal menstrual cycle, rising levels of FSH stimulate the emergence of a dominant ovarian follicle and oestrogen production. When a critical oestrogen level is reached, GnRH pulses trigger the LH surge and ovulation. After ovulation, a corpus luteum is formed which produces progesterone to prepare the endometrium for implantation of a fertilized ovum. After approximately 14 days, if implantation has not occurred, the corpus luteum regresses and progesterone

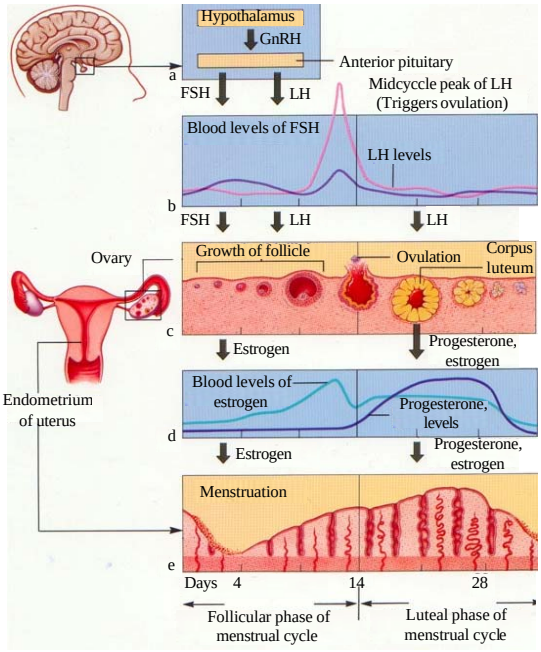

Fig. 1: Hormonal control of the menstrual cycle. Reproduced with permission from http://dentistryandmedicine.blogspot.com/search/label/Gynecology

levels drop. The endometrium can no longer be sustained and menstrual shedding occurs. The spiral arterioles linking the functional and basal layers of the endometrium are able to clamp down causing the menstrual bleeding to stop usually within 7 days.

The hypothalamus is exquisitely sensitive to circulating levels of oestrogen which inhibit GnRH secretion by the hypothalamus (the "negative feedback loop"). This negative feedback loop is present in the foetus and is active throughout childhood and adolescence. However, there is also the "positive feedback loop" where a critical level of oestrogen stimulates pulses of GnRH which triggers the LH surge and ovulation. The latter only develops later in puberty (Golden and Carlson, 2008).

Any disruption in this axis may result in amenorrhoea or menstrual cycle disturbances (Popat *et al*., 2008). The correct functioning of other endocrine glands (thyroid, adrenal cortex and pancreas) is also crucial for correct reproductive function (Petraglia *et al*., 2008).

Definition of amenorrhoea: Amenorrhoea is the absence of menses and may be primary or secondary. Primary amenorrhoea is the absence of menstrual bleeding and secondary sexual characteristics in a female by age 14 years or the absence of menstrual bleeding with normal development of secondary sexual characteristics in a female by age 16 years. Secondary amenorrhoea is more common and is the absence of menstrual bleeding in a woman who had been menstruating but later stops menstruating for 3 or more months in the absence of pregnancy, lactation, cycle suppression with systemic hormonal contraceptive pills or menopause (Golden and Carlson, 2008). Secondary amenorrhoea is commoner than primary amenorrhoea (Speroff and Fritz, 2005). The causes of amenorrhoea can be endocrine, genetic or anatomic (Fig. 2).

Functional Hypothalamic Amenorrhoea (FHA) (also known as hypogonadotropic hypogonadism) is defined as a non-organic and reversible disorder in which the impairment of Gonadotropin-Releasing Hormone (GnRH) pulsatile secretion plays a key role (Yen, 1998). It is the most common cause of secondary amenorrhoea, being responsible for approximately 35% of cases (Marshall, 1994). There are main three types of FHA: stress-related amenorrhoea, weight loss-related amenorrhoea and exercise-related amenorrhoea (Meczekalski *et al*., 2008).

HYPOTHALAMIC (FUNCTIONAL) AMENORRHOEA

Common features associated with individuals at risk for this disorder can include: being thin or underweight; having higher levels of perfectionism; engaging in athletic or intellectual pursuits; reporting adverse childhood experiences such as sexual molestation; exposure to increased stress; need for social approval and altered attitudes towards eating. These features overlap with those of individuals presenting with eating disorders such as anorexia nervosa and bulimia and those presenting with exercise-associated amenorrhoea. These personality

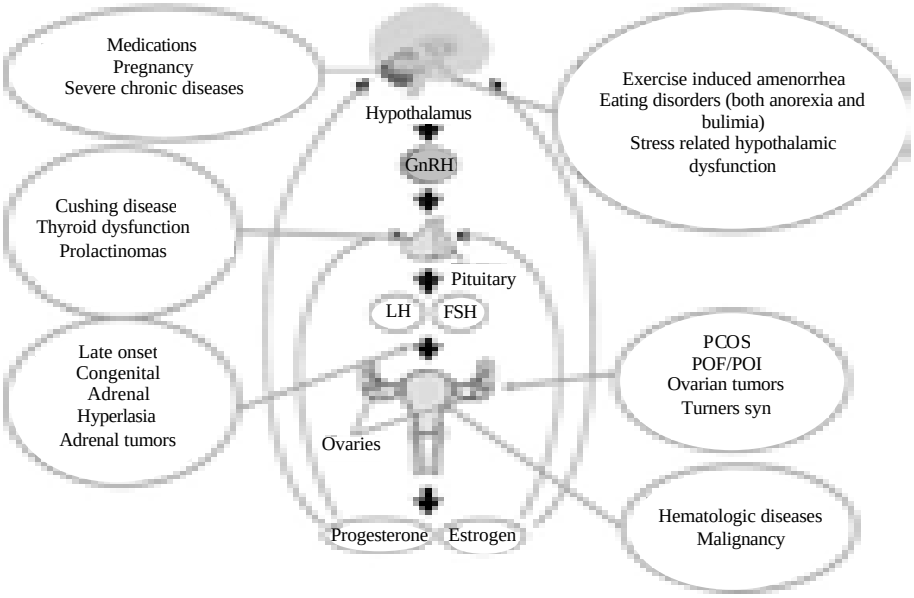

Fig. 2: Conditions associated with amenorrhoea. Reproduced with permission from (Popat *et al*., 2008)

characteristics indicate that a thorough psychological evaluation and medical history is an essential part of the clinical evaluation in these young women.

Patients with hypothalamic amenorrhoea are categorized by low or normal gonadotropins, normal prolactin levels, a normal imaging evaluation of the sella turcica and a failure to demonstrate withdrawal bleeding. A progestin challenge test will usually result in scant or no menstrual bleeding; however addition of combined oestrogen with progestin will result in endometrial growth followed by menses because the uterine compartment remains functionally normal.

In addition to the history and the exclusion of pregnancy, a thorough evaluation of individuals presenting with HA will require baseline studies of FSH prolactin and oestradiol. A good practice is to evaluate such patients annually. This annual surveillance should include a prolactin assay and the coned-down view of the sella turcica. The X-ray is necessary only every 2-3 years after several years with no change (Speroff *et al*., 1999).

The discovery of leptin has given us insight into the mechanisms of hypothalamic amenorrhea. Leptin is a protein produced by the adipocyte which acts on the hypothalamus to regulate food intake, energy expenditure and body weight. In addition it also plays a role in sexual maturation and reproductive functioning. The current thought is that in both anorexia nervosa and in exercise-induced amenorrhea, amenorrhea is an adaptive response to an energy deficit, mediated in part by leptin. Leptin levels in are low in anorexia nervosa and increase with refeeding (Eckert *et al*., 1998; Grinspoon *et al*., 1996; Haas *et al*., 2005; Misva *et al*., 2004).

Administration of recombinant leptin to women with hypothalamic amenorrhoea has been shown to restore LH pulsatility and ovulatory menstrual cycles (Chou *et al*., 2011; Welt *et al*., 2004).

Eating disorders: The prevalence of anorexia nervosa in young women is 0.3-0.5% with the highest incidence in adolescent girls between the ages of 15 and 19 years. While amenorrhoea is one of the features necessary for the diagnosis of anorexia nervosa, dietary restraint even in the presence of a normal body weight can lead to hypothalamic amenorrhoea.

Amenorrhoea in anorexia nervosa is associated with a marked reduction in gonadotropin secretion to a level comparable with prepubertal girls. The underlying cause of this hypogonadotropic state is the reduced frequency and amplitude of LH pulsatile release due to the arrest of hypothalamic GnRH pulsatile activity. Oestradiol secretion is low while the hypothalamic Corticotrophin Releasing Hormone (CRH) adrenocorticotrophic hormone adrenal axis is activated by hypersecretion of cortisol. CRH inhibits hypothalamic GnRH secretion and glucocorticoids inhibit pituitary LH and ovarian oestrogen and progesterone secretion. Insulin sensitivity is increased, glucose effectiveness is reduced and fasting plasma glucose and insulin levels are low (Petraglia *et al*., 2008).

Endocrine studies can thus be summarized as follows: FSH and LH levels are low, cortisol levels are elevated, prolactin levels are normal, TSH and thyroxine levels are normal but the 3, 5, 3 triiodotyronine level is low and reverse T3 is high. Indeed, many of the symptoms can be attributed to the relative hypothyroidism. There appears to be compensation to the state of undernourishment with diversion from formation of the active T3 to the inactive metabolite, reverse T3. With weight gain, all of the metabolic changes revert to normal.

Clinically a spectrum is encountered that ranges from a period of amenorrhoea to the severely ill patient. Besides amenorrhoea, constipation is a common symptom, often severe and accompanied by abdominal pain. Hypotension, hypothermia, rough dry skin, soft lanugo-type hair on the back and buttocks, bradycardia and oedema are the most commonly encountered signs. An elevation of serum carotene is not always associated with a large intake of yellow vegetables, suggesting that a defect of vitamin A utilization is present (Kins, 1989).

A careful and gentle revelation to the patient of the relationship between the amenorrhoea and low body weight is often all that is necessary. After 1 year of treatment, approximately two-thirds of patients resume menses and after 2 years, approximately 95% do so. If progress is slow, hormone therapy should be initiated and if there is continued weight loss, psychiatric and/or psychological consultation should be considered.

The prevalence of bulimia nervosa in young women is approximately 5%. In contrast to anorexia nervosa, patients with bulimia nervosa are usually of normal weight and usually have regular menses. However, they may also have irregular menses as a result of repeated episodes of dietary restriction. They do not usually have prolonged amenorrhoea though, unless they are also of low weight in which case, they would meet diagnostic criteria for anorexia nervosa, binge eating/purging type (Katzman and Golden, 2007).

Exercise-induced amenorrhoea: Generally, exercise-induced amenorrhoea results from suppression of the GnRH pulsatility leading to hypooestrogenism. Athletes may be consuming too few calories to meet their energy needs thus resulting in a state of chronic caloric deprivation (Loucks *et al*., 1998). They may also be underweight. In sports that emphasize leanness, amenorrhoea results from hypo-oestrogenism secondary to suppression of the HPO axis. However, in sports where

Fig. 3: Extreme stress can lead to amenorrhoea. Reproduced with permission from http://www.souhridam.com/stress-help-center/

strength rather than low body weight is the emphasis, another mechanism may explain the hypothalamic disruption of the reproductive axis. These athletes often have elevated androgen levels, elevated LH levels and elevated LH/FSH ratios. It has been hypothesized that the elevated androgens are secondary to stimulation of the hypothalamic-pituitary-adrenal axis. This increase in androgen levels may impair follicular development at the ovarian level thus causing anovulation and amenorrhoea (Constantini and Warren, 1995).

Stress-induced amenorrhoea: Physical and psychosocial stress (Fig. 3) interrupts homeostasis and redirects energy and other resources away from nonessential functions such as reproduction, to the central nervous system and cardiovascular systems. There is activation of the hypothalamic-pituitary-adrenal axis with increased secretion of Corticotropin Releasing Hormone (CRH) and stimulation of the sympathetic nervous system with release of epinephrine and norepinephrine. Secretion of CRH is responsible for activation of central endogenous opioid activity.

The HPO axis is inhibited at a number of levels by complex interrelated mechanisms. Both CRH and endogenous opioids directly inhibit GnRH release by the hypothalamus. In addition, glucocorticoids inhibit pituitary LH secretion as well as ovarian oestrogen and progesterone production (Magiakou *et al*., 1997).

Chronic illness: Chronic illness can lead to pubertal delay and cessation of menses. For example, renal disease, liver disease, immunodeficiencies, inflammatory bowel disease and uncontrolled diabetes have been associated with anovulation and amenorrhoea (Golden and Carlson, 2008).

Malnutrition may be a leading factor in causing amenorrhoea. Many diseases cause an increased caloric need: intestinal diseases leading to malabsorption and protein wasting, the chronic inflammation of juvenile arthritis and increased cardiac expenditures in congenital heart disease. Despite the increased demand, many patients have decreased intake either from suppressed appetite or from a concomitant eating disorder that is found with increased prevalence in certain diseases such as type 1 diabetes mellitus. The resulting caloric discrepancy may lead to chronic malnutrition, leading to amenorrhoea (Rosmark *et al*., 1986; Jones *et al*., 2000; Rydall *et al*., 1997).

Medication-induced amenorrhoea: Amenorrhoea is also seen in patients taking antipsychotics or antidepressants. In fact, approximately 50% of patients who are taking antipsychotic medication develop menstrual disturbances and 12% develop amenorrhoea. Many of the antipsychotic drugs block pituitary dopamine D2 receptors, thereby removing the inhibitory effect of dopamine on prolactin secretion by the pituitary. Prolactin levels can increase five to tenfold (Wieck and Haddad, 2003).

Antidepressants with increased serotonergic activity (e.g., the selective serotonin receptor inhibitors, the monoamine oxidase inhibitors and some tricyclics) may also increase prolactin levels but to a lesser degree. Discontinuing the offending drug usually results in resolution of the problem. Another group of medications frequently used in the adolescent age group and that can cause amenorrhoea is injectable contraceptives (Petraglia *et al*., 2008). Amenorrhoea can rarely follow the discontinuation of oral contraceptives what is known as "post-pill amenorrhoea".

Kallmann syndrome: Kallmann syndrome is a genetic disorder caused by one or more mutations of the *KAL* gene at Xp22.3.48. It is the association of isolated hypogonadotropic hypogonadism with anosmia. The syndrome affects approximately 1 in 10,000 males and 1 in 50,000 females. In this syndrome, there is a defect in the process of migration of the olfactory and GnRH neurons. Girls with Kallmann syndrome typically are not identified until adolescence when they present with failure of sexual development and primary amenorrhoea (Golden and Carlson, 2008).

Hyperprolactinaemia: Hyperprolactinaemia is a common condition with varied aetiology (Zargar *et al*., 2004). In which differential diagnosis of hyperprolactinaemia adapted from (Aron *et al*., 1995):

Physiologic
Pregnancy, lactation
Nonphysiologic
Prolactin-secreting tumours
  Prolactinomas-unihormonal
  Tumours secreting multiple hormones
Drugs
  Dopamine synthesis inhibitors, depletors and receptor blockers-Alpha-methyldopoa, reserpine, verapamil, phenothiazines, Thiothixenes and butyrophenones
  Others-oestrogen, narcotics
Central nervous system disorders that lead to pituitary disinhibition
  Hypothalamic lesions-tumour, sarcoid, histiocytosis X and infiltrative diseases
  Pituitary stalk lesions-trauma (stalk section) and compression by tumour or other mass lesions
Miscellaneous
  Systemic illness cirrhosis, renal failure
  Primary hypothyroidism
  Chest wall and spinal cord lesions-postsurgical, herpes zoster, burns
  Polycystic ovary syndrome
  Macroprolactinaemia
  Idiopathic

Women may present with decreased libido, infertility, oligomenorrhea/amenorrhoea and galactorrhoea (Luciano, 1999). The prevalence of hyperprolactinemia is as high as 20% in women with unexplained primary or secondary amenorrhoea, even when galactorrhoea or other symptoms of pituitary dysfunction are not present. Hyperprolactinemia's effect on the menstrual status is via its effect on the pulsatility of GnRH. With interference of the normal GnRH pulsatility, gonadotropins are suppressed and a subsequent decrease in oestrogen levels is seen.

Multiple causes of hyperprolactinemia exist falling into three general categories: physiologic, pharmacologic and pathologic. They include physiological states such as pregnancy and lactation, certain medications such as neuroleptics, endocrinopathies such as primary hypothyroidism and PCOS, systemic diseases such as systemic lupus erythematosus or rheumatoid arthritis and chronic renal failure. When specific treatable underlying causes have been eliminated and in cases of severe hyperprolactinemia, the most likely cause is a Prolactin (PRL) secreting pituitary adenoma (Biller, 1999).

An initial level of prolactin above the normal range should be followed by a repeat level from a blood sample drawn in the morning with the patient in a fasting state. The medical history and a few laboratory tests can eliminate the most common physiologic and pharmacologic causes of hyperprolactinemia including pregnancy, primary hypothyroidism and treatment with drugs that reduce dopaminergic effects on the pituitary. In the absence of such causes, radiologic imaging of the sella turcica is necessary to establish whether a PRL-secreting pituitary adenoma or other lesion is present. The vast majority of patients with a pituitary adenoma are treated medically with dopamine agonist drugs. Surgery is reserved for the patient with the uncommon tumour that does not respond to medical therapy or has a large cystic component or for the occasional patient who cannot tolerate dopamine agonists or who experiences pituitary apoplexy (Biller *et al*., 1999).

Prolactinomas: Prolactinomas are the most common functioning pituitary tumour. The 90% are intrasellar adenomas that rarely increase in size. The rest are macroadenomas that usually come to clinical attention because of local mass effects. In women, hypersecretion of prolactin leads to amenorrhoea, galactorrhoea and infertility. The amenorrhoea is usually secondary and may follow pregnancy or the use of oral contraceptives. Hyperprolactinemia may lead to bone loss due to the inhibitory effect of prolactin on sex steroids.

Pharmacological intervention should be considered the first line therapy and involves the use of dopamine agonists to reduce tumour size and prolactin levels. Bromocriptine has the longest history of use and is a well-established, inexpensive, safe and effective therapy option. There does not appear to be any increased risk of abortion, malformations or multiple births in pregnancies achieved with bromocriptine thus this dopamine agonist can be used safely during pregnancy. However, bromocriptine requires multiple daily dosing and some patients are resistant or intolerant to this therapy. The two newer dopamine agonists, quinagolide and cabergoline, provide more effective and better tolerated treatments compared with bromocriptine and may offer effective therapies for bromocriptine-resistant or intolerant patients (Crosignani, 2006).

For patients with hyperprolactinaemia, pregnancy is safe and can frequently be beneficial inducing a decrease in prolactin levels. Surgery should be considered only in certain circumstances and for the majority of patients, dopamine agonists will be sufficient to alleviate symptoms and restore normal prolactin levels (Crosignani, 2006). Transsphenoidal neurosurgery achieves immediate resolution of hyperprolactinaemia with resumption of menses in approximately 30% of patients with macroadenomas and 70% of patients with microadenomas. Besides an inability to achieve a complete cure, surgery may be followed by recurrence of tumour. Other complications of surgery include cerebrospinal fluid leaks, post-op diabetes insipidus and occasionally meningitis (Schlechte *et al*., 1986; Parl *et al*., 1986). Results with radiation therapy are less satisfactory than with surgery. Panhypopituitarism can occur as long as 10 years after treatment. Irradiation should be reserved as adjunctive

therapy for controlling postoperative persistence or regrowth of large tumours and shrinking large tumours that are unresponsive to medical treatment (Snyder *et al*., 1986; Feigenbaum *et al*., 1996).

Craniopharyngioma: Lesions that reside in the hypothalamic/pituitary junction such as craniopharyngioma, Rathke's cleft cyst or rarely gliomas, meningiomas or chordomas may result in menstrual dysfunction. Craniopharyngiomas can present with amenorrhoea and/or hyperprolactinaemia. They are epithelial tumours arising from the craniopharyngeal duct in the sellar or parasellar region. They represent 2-5% of all intracranial neoplasms. They have an uncertain pathogenesis and present with a variety of symptoms-endocrine, visual, behavioural and cognitive. Hormonal disruption is prevalent and a deficiency of LH/FSH is found in approximately 38-82% of patients. This disruption is the likely mechanism for menstrual dysfunction. Another possible mechanism of hyperprolactinemia in patients with craniopharyngioma is direct encroachment on the hypothalamus and/or pituitary stalk by the tumour and interruption of prolactin inhibiting factor.

HYPOPITUITARISM

Hypopituitarism may cause amenorrhoea. Both genetic and acquired disorders can cause hypopituitarism. These include molecular mutations that result in one or more congenital pituitary hormone defects, pituitary and hypothalamic tumours, infiltrative or inflammatory disease and traumatic brain injury. The incidence of hypopituitarism is 12-42 new cases per million per year.

These patients present with the typical signs and symptoms of hypogonadism in addition to the symptoms and signs of other associated pituitary hormonal insufficiencies. Gonadotropins (FSH and LH) and sex steroid hormone (oestrogen and progesterone) levels are low. Other pituitary hormone deficiencies require dynamic testing for their diagnosis, however, testing for pituitary gonadotropin deficiency relies mainly on history, physical exam and baseline laboratory measurement of FSH, LH and oestradiol.

When pituitary hormone insufficiency occurs due to tumour compression of normal pituitary hormone production, growth hormone is often the first hormone to be deficient then LH and FSH and later TSH and ACTH. Infrequently, metastasis from other tumours such as breast, lung or renal can result in hypogonadism associated with other hormonal deficiencies. Metastasis will more often be associated with posterior hormone abnormalities such as diabetes insipidus whereas primary pituitary tumours will usually have only anterior hormone dysfunction.

Hemachromotosis, due to selective iron deposition in the gonadotropes can cause hypopituitarism. Other infiltrative disorders of the pituitary include granulomatous diseases such as sarcoidosis, tuberculosis and histiocytosis X. Lymphocytic hypophysitis is due to a lymphocytic infiltration of the pituitary, usually present in patients during pregnancy or post partum, although it can occur at other times. It is associated with other pituitary hormonal dysfunction and the timing of the loss of gonadotrope function is later with ACTH and TSH affected earlier, than generally observed in pituitary adenomas. More recently traumatic brain injury is being recognized as a cause of hypopituitarism.

Single or multiple pituitary deficits have been documented in up to two-thirds of affected patients with TB. The HPO and growth hormone axes are the most commonly affected (Rothman and Wierman, 2008).

Sheehans's syndrome (also known as postpartum hypopituitarism or postpartum pituitary necrosis) is hypopituitarism caused by necrosis due to blood loss and hypovolemic shock during and after childbirth. Symptoms of Sheehan syndrome may include inability to breast-feed, fatigue, amenorrhoea or oligomenorrhoea, loss of pubic and axillary hair and hypotension. Symptoms other than difficulty with breastfeeding may not develop for several years after the delivery (Schrager and Sabo, 2001).

OVARIAN DISORDERS

Polycystic Ovary Syndrome (PCOS): Polycystic Ovary Syndrome (PCOS) is one of the most common human endocrinopathies, affecting 5-10% of women of reproductive age. It is the most common cause of hyperandrogenic chronic anovulation. The primary etiology of PCOS is unknown but resistance to insulin is thought to be a fundamental component.

The diagnosis of PCOS was previously based on a combination of clinical and endocrine features including raised serum concentrations of Luteinizing Hormone (LH), Testosterone (T) and androstenedione and reduced levels of sex hormone binding globulin. The diagnostic criteria for PCOS are ovarian dysfunction evidenced by oligomenorrhea or amenorrhoea and clinical evidence of androgen excess (e.g., hirsutism and acne) in the absence of other conditions that can cause these same symptoms (King, 2006). With the introduction of pelvic ultrasound in the 1980s, non-invasive assessment of ovarian morphology became possible. This gives a definite diagnosis.

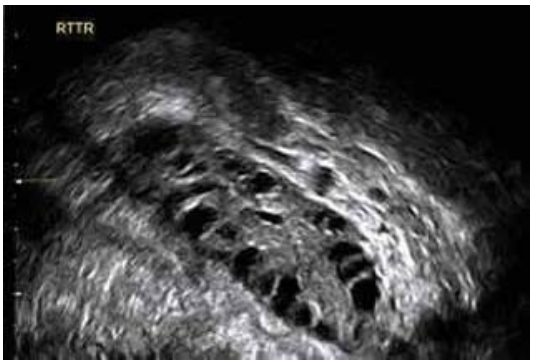

Fig. 4: Transvaginal image of a polycystic ovary showing peripheral distribution of follicles. Reproduced with permission from http://www.advancedfertility.com/pcos.htm

In fact, transvaginal ultrasound is currently the gold standard for diagnosing polycystic ovaries (Fig. 4). The ultrasound criteria for diagnosing PCOS have evolved from simply increased dimensions to the recognition of a characteristic follicular pattern and textural changes in the ovarian stroma (Lakhani *et al*., 2002).

PCOS underlies irregular menses in up to one-third of girls (Venturoli *et al*., 1987). Menarche is not usually delayed but bleeding is then persistently irregular. In adolescents, PCOS can present with primary or secondary amenorrhoea, acne, hirsutism or merely irregular periods. Menstrual disturbance is likely to be the main issue for adolescents with PCOS but the established long-term risks of obesity, subfertility and diabetes as well as the possible risks of endometrial hyperplasia (Hardiman *et al*., 2003) and carcinoma and cardiovascular disease (Rajkowha *et al*., 2000) and breast cancer (Balen, 2001) require consideration. Patients with PCOS have excess unopposed circulating oestrogen, increasing their risk of endometrial cancer three fold. The insulin resistance associated with PCOS increases a patient's risk of diabetes mellitus two to five fold; therefore, testing for glucose intolerance should be considered.

The goals of treatment should focus on restoring menstrual regularity, decreasing androgen excesses and decreasing insulin resistance (Hoyt and Schmidt, 2004). The primary treatment for PCOS is weight loss through diet and exercise. Modest weight loss can lower androgen levels, improve hirsutism, normalize menses and decrease insulin resistance. It may take months to see these results however. Use of oral contraceptive pills or cyclic progestational agents can help maintain a normal endometrium.

Hyperandrogenism and hirsutism are distressing symptoms for young women. Optimally treatment combines cosmetic and medical therapies. Medical regimens stop further progression of hirsutism and slow the rate of hair growth. However, drug therapies may take 6±9 months or longer before any benefit is perceived and so laser, electrolysis, waxing and bleaching may be helpful in the interim.

Hyperandrogenism can be treated by a combination of an oestrogen (such as ethinyloestradiol or a combined contraceptive pill) and the anti-androgen Cyproterone Acetate (CPA, 50±100 mg). Oestrogens lower circulating androgens by a combination of a slight inhibition of gonadotropin secretion and gonadotropin-sensitive ovarian steroid production and by an increase in hepatic production of sex hormone-binding globulin resulting in lower free testosterone. The CPA is taken for the 1st 10 days of a cycle (the 'Reversed Sequential' Method) and the oestrogen for the 1st 21 days. After a gap of exactly 7 days, during which menstruation usually occurs, the regimen is repeated.

Spironolactone, a potassium sparing diuretic has antiandrogenic properties and is useful in women for whom the oral contraceptive pill is contra-indicated (e.g., because of hypertension). Spironolactone, at a dose of 50±200 mg daily may result in erratic menstrual bleeding and should be combined with reliable contraception. A new COCP, Yasmina (Shering UK), contains the progestogen, drospirenone which is a derivative of spironolactone with potential anti-androgenic properties and benefits for women with PCOS.

Although, diet is the first line treatment for improving insulin sensitivity in overweight adolescents with PCOS, insulin sensitizing agents such as metformin are becoming increasingly popular in the management of PCOS as they act directly on insulin resistance and help correct both metabolic and endocrine problems (Hickey and Balen, 2003).

PREMATURE OVARIAN FAILURE

Premature Ovarian Failure (POF) is the occurrence of hypergonadotropic hypooestrogenic amenorrhoea in women under the age of 40 years. It occurs in 1% of women. POF is idiopathic in 74-90% of cases but can be familial (4-33%) or sporadic.

The known causes include: genetic aberrations which could involve the X chromosome or autosomes. Autoimmune ovarian damage as POF is associated with other autoimmune disorders. Iatrogenic following surgical, radiotherapeutic or chemotherapeutic interventions. Environmental factors like viral infections and toxins (Goswami and Conway, 2005).

As in the case of physiological menopause, POF presents by typical manifestations of climacterium: infertility associated with palpitations, heat intolerance, flushes, anxiety, depression and fatigue. Besides infertility, hormone defects may cause severe neurological, metabolic or cardiovascular consequences and lead to the early onset of osteoporosis.

POF is biochemically characterized by low levels of gonadal hormones (oestrogens and inhibins) and high levels of gonadotropins (LH and FSH) (hypergonadotropic amenorrhoea). The diagnosis is based on finding of amenorrhoea before age 40 associated with FSH levels in the menopausal range (high serum FSH concentrations (140 IU L^{-1}) on at least two occasions separated by a few weeks) (Goswami and Conway, 2007). The reason for the need for two samples is that the diagnosis is often devastating and certainty is required and also because the natural history of POF can be very variable.

Screening for associated autoimmune disorders and karyotyping, particularly in early onset disease, constitute part of the diagnostic work-up. There is no role of ovarian biopsy or ultrasound in making the diagnosis. Affected women should be investigated for premutations of the *FMR1* gene (causing fragile X syndrome in its fullest form) and for adrenal antibodies. Thyroiditis is the most frequent autoimmune disorder associated with premature ovarian failure and should be ruled out as well.

Management essentially involves hormone replacement and infertility treatment, the only proven means for the latter being assisted conception with donated oocytes. Long-term HRT is needed for relief of menopausal symptoms and to prevent long-term health sequel of oestrogen deficiency such as osteoporosis (Davis, 1996). Oestrogen replacement is usually continued up to the age of 50 years when the risk and benefit of continued treatment are reviewed (Goswami and Conway, 2007).

A careful family history can identify other affected female members in as many as 30% of cases whose relatives can then be offered genetic counselling (Van Kasteren *et al*., 1999).

THYROID DISORDERS

Both hypo and hyperthyroidism can have effects on the HPO axis and menstrual irregularities are more common in this population when compared with healthy controls. Patients with hypothyroidism may present with heavy or irregular menses as there are also direct effects on clotting factors influencing bleeding patterns. Patients with hyperthyroidism will have shortened anovulatory cycles that can result in infertility. GnRH secretion patterns and sex hormone binding globulin levels are also directly affected by thyroid disorders but random LH, FSH and E2 levels are often non-diagnostic (Rothman and Wierman, 2008).

Thyroid disease is more prevalent in females than males and often presents in adolescence. Menstrual disturbances in hypothyroid patients range from 20-70%. Although, hypermenorrhoea or oligomenorrhoea are the more prevalent menstrual disorders, amenorrhoea can also be seen. One proposed mechanism for the amenorrhoea is through the effect of Thyrotopin-Releasing Hormone (TRH) on prolactin levels. This releasing hormone acts on the thyrotrophs to release TSH and on lactotrophs to release prolactin. Secondary to the increased levels of TSH-releasing hormone found in hypothyroidism increased levels of prolactin are released, leading to functional hyperprolactinemia (Koutras, 1997). The estimated incidence of hyperprolactinaemia in hypothyroidism ranges from 0-40% (Raber *et al*., 2003).

Patients with hyperthyroidism have rates of menstrual irregularities ranging from 20-60% with rates of amenorrhoea reaching up to 20%. The exact mechanism for the amenorrhoea is not clear but appears to be a combination of hormonal abnormalities, nutritional deficiencies and emotional stress that may accompany hyperthyroidism. Hyperthyroidism results in increased levels of sex hormone-binding globulins which may cause an increased level of plasma oestrogen. Levels of circulating androgens, particularly testosterone and androstenedione are also elevated as is their conversion to their oestrogen counterpart, oestradiol or estrone (Krassas, 2000). While mean LH levels are generally higher in hyperthyroid women, LH peaks may be completely absent in women with amenorrhoea (Golden and Carlson, 2008).

ADRENAL CAUSES OF AMENORRHOEA

Congenital adrenal hyperplasia: Congenital adrenal hyperplasia refers to a group of autosomal recessive disorders of steroidogenesis. A number of enzyme deficiencies have been found but >90% of cases are caused by deficiency of 21-hydroxylase which catalyses the conversion of progesterone to deoxycorticosterone and 17-OH progesterone to 11-deoxycortisol. There is an interruption in the pathway leading to synthesis of mineralocorticoids and glucocorticoids. Consequently, progesterone, 17-OH progesterone and its precursors are, instead, shunted to the androgen pathway. Hypothalamic secretion of CRH increases to overcome the effects of the block. As a result, ACTH secretion is increased, further increasing androgen production. The two other major enzyme deficiencies are deficiencies of 11-beta-hydroxylase and 3-beta-hydroxysteroid dehydrogenase.

There are two major clinical forms of congenital adrenal hyperplasia, depending on whether the enzyme deficiency is complete or partial the classic form and the nonclassic form. The classic form usually presents in infancy with salt wasting or ambiguous genitalia. It occurs in approximately one in 16,000 births (Spesier and White, 2003). The nonclassic form is one of the most frequently seen autosomal genetic disorders and occurs in approximately 0.2% of the general population. This nonclassic form of congenital adrenal hyperplasia usually presents in childhood and is characterized by premature pubarche and in adolescence by hirsutism or amenorrhoea (Moran *et al*., 2000). Hyperandrogenism has a direct effect on the hypothalamus, suppressing pituitary-ovarian function. The suppression of the HPO axis can be reversed after adequate treatment of the excess adrenal androgen production (Klingensmith *et al*., 1976).

Cushing's syndrome: Cushing's syndrome is caused by high circulating levels of cortisol. In adolescents, Cushing's syndrome is most frequently caused by iatrogenic exogenous administration of corticosteroids. Other causes include hypersecretion of corticotropin by a microadenoma of the anterior pituitary (Cushing's disease); secretion of corticotropin by an adrenal tumour or, occasionally, ectopic production of corticotropin by a nonpituitary tumour such as carcinoma of the lung. The clinical findings are usually self-evident. Oligomenorrhoea and amenorrhoea may be part of the clinical picture. The pathophysiology is direct suppression of the HPO axis (Golden and Carlson, 2008).

EVALUATION OF AMENORRHOEA

Evaluation of menstrual cycle irregularity depends on a careful assessment of the history, physical examination and various laboratory and diagnostic tests (Master-Hunter and Heiman, 2006). This should be done systematically according to the suspected origin of the dysfunction (i.e., outflow tract, ovary or hypothalamus/pituitary). In cases of secondary amenorrhoea, however, unless organic disease is suspected or the woman is desperately seeking relief from infertility, most experts would not investigate until the amenorrhoea has lasted for 6-12 months as most women start menstruating during this time (Oats and Abraham, 2005).

History: As part of the history (Fig. 5), one should ask first ask about the possibility of pregnancy as this is the most common cause of amenorrhoea. Determining whether the patient is sexually active and whether she is using contraceptive methods is important. In some cases, the hormonal contraception itself may be the cause of the amenorrhoea. Unusually thin patients should be asked questions about exercise, weight gain/loss and eating habits. One should also ask about associated symptoms like galactorrhoea, headache, reduced peripheral vision, hirsutism, vaginal dryness, hot flushes, night sweats, weight gain or weight loss or excessive anxiety (Morrison, 1998).

Fig. 5: A good history is imperative. Reproduced with permission from http://www.sciencephoto.com/media/298794/enlarge

Inquiring about other aspects of growth and pubertal development is important. Breast development, pubertal growth spurt and adrenarche are delayed or absent in persons with hypothalamic pituitary failure. A distinguishing factor in the case of isolated ovarian insufficiency or failure is that adrenarche occurs normally while oestrogen-dependent breast development and the pubertal growth spurt are absent or delayed.

Current medication (s), sexual activity use of over-the-counter preparations, contraception, current and past medical conditions, surgery and any history of illicit and prescribed drug use are generic to primary and secondary amenorrhoea. Taking a full drug history is imperative as many drugs including antidepressants, neuroleptics and the oral contraceptive pill can cause amenorrhoea. Also, abuse of drugs such as cocaine and opioids have central nervous system effects that may disrupt the menstrual cycle.

Taking a full family history is also important. One should ask about autoimmune disorders in the family. Those with primary amenorrhoea need to be queried as to mother's age at menarche and any known or suspected congenital disorders. Patients with secondary

amenorrhoea are questioned about mother's age at menopause, any pregnancies (including complications) and their menstrual cycle (Wilson *et al*., 2005).

A history of otherwise normal growth and pubertal development and cyclic pelvic pain in association with primary amenorrhoea suggests the possibility of a congenital outflow tract abnormality such as imperforate hymen or agenesis of the vagina, cervix or uterus. Prior history of a surgical procedure involving the endometrial cavity especially if performed in the presence of infection, raises the possibility of uterine synechiae (Asherman's syndrome). History of minimal body hair, good breast development with blind vagina and absent uterus is suggestive of androgen resistance syndrome. Symptoms of vaginal dryness, hot flashes, night sweats or disordered sleep may be a sign of primary ovarian insufficiency or premature ovarian failure. Prior history of chemotherapy or radiation therapy may be associated with primary ovarian insufficiency.

Associated galactorrhoea, headaches or reduced peripheral vision could be a sign of intracranial tumour such as prolactinoma. These symptoms require immediate further evaluation. An impaired sense of smell in association with primary amenorrhoea and failure of normal pubertal development may be related to isolated gonadotropin deficiency as is observed in persons with Kallmann syndrome. Sarcoidosis can manifest insidiously, with development of mild fatigue, malaise, anorexia, weight loss and fever. Cough and dyspnea may also be present. Hemachromatosis may manifest as weakness, lassitude, weight loss and a change in skin colour.

Autoimmune adrenal insufficiency, a potentially fatal condition, often manifests as vague and nonspecific symptoms. Amenorrhoea may be the first clear symptom indicating a need for further evaluation to detect this condition. Amenorrhoea may herald the onset of other autoimmune endocrine disorders such as hyperthyroidism, hypothyroidism or autoimmune lymphocytic hypophysitis. The same is true for other endocrine disorders such as cushing syndrome or pheochromocytoma. A careful review of symptoms may help uncover these disorders.

AIDS, HIV disease or other types of immunodeficiency states may induce systemic infection, leading to chronic disease and amenorrhoea. Occult malignancy with progressive weight loss and a catabolic state may lead to loss of menstrual regularity. A careful review of systems may help uncover such a disorder.

Physical examination: Physical examination should begin with observation and an overall assessment of nutritional status and general health. For the patient with primary amenorrhea, the physical examination should focus on pubertal development and possible genital outflow obstruction. For patients with secondary amenorrhea, the physical examination should focus on signs of hyperandrogenism and insulin resistance as well as evidence of weight loss. Height and weight should be measured and evidence sought for chronic disease or cachexia (Golden and Carlson, 2008). Hypothermia, bradycardia, hypotension and reduced subcutaneous fat can be observed in persons with severe anorexia nervosa. In cases of frequent vomiting, look for possible dental erosion, reduced gag reflex, trauma to the palate, subconjunctival hemorrhage and metacarpophalangeal calluses or bruises. Skin examination findings can also give clues to other endocrine disorders. Vitiligo or increased pigmentation of the palmar creases may herald primary adrenal insufficiency. Thin, parchment-like skin, striae and evidence of easy bruising may be signs of Cushing syndrome. Warm, moist skin radiating excessive heat may be a sign of hyperthyroidism. Large pituitary tumours can cause visual-field cuts by impinging on the optic tract. In some cases, these visual-field cuts can be detected by simple confrontational testing.

One should seek for the presence or absence of secondary sexual characteristics. The skin should also be examined for evidence of androgen excess such as hirsutism and acne. Acanthosis nigricans may be present in association with androgen excess related to insulin resistance. The state of breast development and presence of galactorrhoea should be assessed. In some cases, breast discharge can be expressed, yet the condition is not true galactorrhoea. If the discharge is indeed milk, this can be confirmed by finding fat globules in the fluid using low-power microscopy. The presence of axillary and pubic hair should be evaluated. These are a marker of adrenal and ovarian androgen secretion. In cases of panhypopituitarism, sources of androgen are low and pubic and axillary hair is sparse. In addition, some women develop the combination of autoimmune premature ovarian failure and autoimmune primary adrenal insufficiency. These women are also markedly androgen-deficient and have scant axillary and pubic hair. The same is true for persons with androgen insensitivity syndrome (testicular feminization), 17-hydroxylase deficiency and 17, 20 desmolase deficiency.

The abdomen should be examined for any evidence of pregnancy or masses. In cases of primary amenorrhoea with otherwise normal pubertal development, pelvic examination may help detect imperforate hymen, a transverse vaginal septum or cervical or uterine aplasia. A clinician needs to be extra sensitive about pelvic exam in an adolescent who may be dealing with many unknowns

about her health. In many instances, this could be her first pelvic exam. Pelvic examination findings can provide physical evidence indicating the adequacy of oestrogen production. Thin and pale vaginal mucosa with absent rugae is evidence of oestrogen deficiency. Ovarian enlargement may be found upon pelvic examination in cases of autoimmune oophoritis, 17-hydroxylase deficiency or 17, 20 desmolase deficiency. In these disorders, inadequate negative feedback supplied by the ovary permits excessive gonadotropin stimulation which may cause ovarian enlargement with multiple follicular cysts. In some cases, these disorders manifest with an acute onset of pain related to ovarian torsion. A general physical examination may undercover unexpected findings that are indirectly related to the loss of menstrual regularity (e.g., hepatosplenomegaly which may lead to detection of a chronic systemic disease).

Anomalies such as short stature, webbed neck, multiple ear infections, coarctation of the aorta, renal abnormalities, hypertension, pigmented nevi, short forth metacarpal and metatarsals, Hashimoto's thyroiditis, obesity and osteoporosis suggest Turner syndrome.

Laboratory studies: A pregnancy test should be the first step. Serum prolactin, FSH, LH and oestradiol levels should be measured routinely in the initial evaluation of amenorrhoea once pregnancy has been excluded (Wilson *et al*., 2005). Prolactin levels in excess of 200 ng mL^{-1} are generally observed only in the case of a prolactinoma. In general, the serum prolactin level correlates with the size of the tumour. Psychotropic drugs, hypothyroidism, stress and meals can also raise prolactin levels. Repeatedly elevated prolactin levels require further evaluation if the cause is not readily apparent. Complete blood cell count, urinalysis and serum chemistries may be indicated to help rule out systemic disease. Thyroid-Stimulating Hormone (TSH) and free thyroxine (T4) should be performed if symptoms or signs of hypothyroidism are present.

An FSH level in the menopausal range is indicative of ovarian insufficiency. If a repeat value in 1 month confirms this finding and amenorrhoea persists, the diagnosis of premature ovarian failure or primary ovarian insufficiency is confirmed. Luteinizing hormone is elevated in cases of 17,20-lyase deficiency, 17-hydroxylase deficiency and premature ovarian failure.

Gonadotropin (FSH and LH) levels should be optimally obtained in early morning on days 1-5 after the onset of menstrual bleeding. In normal women, FSH and LH levels will be similar at this time of the cycle. Women with GnRH deficiency, hypothalamic amenorrhoea and/or pituitary disorders have low or inappropriately normal FSH and LH levels associated with low oestradiol levels. An increased ratio of LH: FSH at this time is often observed in patients with PCOS. Elevated FSH with or without LH and low oestradiol levels is suggestive of primary ovarian failure. In evaluating primary amenorrhoea, both delayed puberty and GnRH deficiency can both present with low FSH and LH. Generally, the passage of time is the only way to distinguish between these two processes.

A testosterone level is often measured to document hyperandrogenism in women with symptoms of acne, hirsutism or frank virilization. Markedly elevated testosterone levels are concerning for but not diagnostic of, hormonally active ovarian or rarely adrenal tumours.

The 24 h measurement of urine free cortisol should be ordered to evaluate for hypercortisolism that may induce menstrual cycle dysfunction. Conversely, a random serum cortisol of 3 Lg dL^{-1} can diagnose adrenal insufficiency however, a patient may have abnormal adrenal function with random cortisol levels within the normal range.

Testing for autoimmune disease is indicated in patients whose FSH, LH and oestrogen levels are consistent with premature ovarian failure (hypergonadotropic type) (Kauffman and Castracane, 2003). Karyotyping is rarely a first-line laboratory test in evaluating amenorrhoea, unless the patient has obvious Turner's stigmata. Karyotyping should be delayed, pending serum androgen levels, in patients suspected of androgen insensitivity. Patients with Mullerian agenesis or gonadal dysgenesis should be karyotyped because chromosomal anomalies are frequently associated with these anomalies (Rattanachaiyanont *et al*., 1997; Arrigo *et al*., 2003).

The progesterone challenge test: Prior to the development of readily available assays to measure serum levels of oestradiol, the progesterone challenge test was used as a bioassay with which to demonstrate oestrogen effect at the level of the endometrium. Progestogens will only provoke bleeding is there is sufficient circulating oestradiol. The test determines if the endometrium will respond and indirectly determines that the oestradiol is above a critical level (Oats and Abraham, 2005). However, the progesterone withdrawal test can provide inappropriately reassuring information that may delay the diagnosis of ovarian insufficiency and possibly other conditions. The progesterone withdrawal test is no substitute for evaluating ovarian health. Demonstrating the presence of normally functioning ovaries requires the concurrent measurement of serum oestradiol and FSH.

Imaging studies and procedures: It is necessary to ascertain presence or absence of vagina, uterus and ovaries in patients with primary amenorrhea. If this cannot be confirmed via physical examination, then trans-abdominal or trans-vaginal ultrasound is used. Data supports the trans-vaginal approach as superior but this may not be an option in a patient with hypoplastic, infantile or virginal vagina. Trans-vaginal ultrasound is also the procedure of choice for evaluating ovaries in suspected PCOS (Khalid, 2004). While more expensive, MRI is a reasonable alternative to ultrasonography in cases not well defined (Reinhold *et al*., 1997).

MRI/CT scan is indicated in the work-up of for pituitary or hypothalamic causes, associated headaches or visual-field cuts, profound oestrogen deficiency with otherwise unexplained amenorrhoea and hyperprolactinemia. Imaging of the hypothalamic/pituitary area is preferable to a study of the entire brain. Hysterosalpingography and hysteroscopy are indicated in cases of possible Asherman's syndrome.

CONCLUSION

Amenorrhoea is a relatively common condition, especially in the adolescent age group and it can cause significant distress. The four most common causes are pregnancy, polycystic ovary syndrome, hypothalamic amenorrhoea, and hyperprolactinaemia. Therefore, the evaluation of amenorrhea always begins with exclusion of pregnancy regardless of whether primary or secondary and irrespective of age. A complete and stepwise history and physical examination are imperative. Further evaluation includes testing Prolactin (PRL), Follicle Stimulating Hormone (FSH), Luteinizing Hormone (LH) and estrogen serum levels. TSH is reserved for selected cases. When history, physical examination and screening laboratory tests do not reveal a cause, a progestin-challenge test may be helpful. Fiber optics, trans-vaginal ultrasonography and Magnetic Resonance (MR) imaging are useful in selected patients. The general principle of treatment is to replace estrogen when hypoestrogenaemia is demonstrated in order to prevent the consequences of long-term estrogen deficiency and osteoporosis. More prolonged deficiency may also increase cardiovascular risk. When a uterus is present, progestogens should also be given to avoid endometrial hyperplasia.

REFERENCES

Aron, D.C., J.B. Tyrell and C.B. Wilson, 1995. Pituitary tumors. Current concepts in diagnosis and management. West J. Med., 162: 340-352.

Arrigo, T., S. Bertelloni, L. Carcione, F. De Luca and C. De Sanctis *et al*., 2003. Characterization of early presentation idiopathic ovarian failure in girls and adolescents. J. Pediatr. Endocrinol. Metab., 16: 835-842.

Balen, A.H., 2001. Polycystic ovary syndrome and cancer. Hum. Reprod. Update, 7: 522-525.

Biller, B.M., 1999. Diagnostic evaluation of hyperprolactinemia. J. Reprod. Med., 44: 1095-1099.

Biller, B.M., A. Luciano, P.G. Crosignani, M. Molitch and D. Olive *et al*., 1999. Guidelines for the diagnosis and treatment of hyperprolactinemia. J. Reprod. Med., 44: 1075-1084.

Chou, S.H., J.P. Chamberland, X. Liu, G. Matarese and C. Gao *et al*., 2011. Leptin is an effective treatment for hypothalamic amenorrhea. Proc. Natl. Acad. Sci., 108: 6585-6590.

Constantini, N.W. and M.P. Warren, 1995. Menstrual dysfunction in swimmers: A distinct entity. J. Clin. Endocrinol. Metab., 80: 2740-2744.

Crosignani, P.G., 2006. Current treatment issues in female hyperprolactinaemia. Eur. J. Obstet. Gynecol. Reprod. Biol., 125: 152-164.

Davis, S.R., 1996. Premature ovarian failure. Maturitas, 23: 1-8.

Eckert, E.D., C. Pomeroy, N. Raymond, P.F. Kohler, P. Thuras and C.Y. Bowers, 1998. Leptin in anorexia nervosa. J. Clin. Endocrinol. Metab., 83: 791-795.

Feigenbaum, S.L., D.E. Downey, C.B. Wilson and R.B. Jaffe, 1996. Transsphenoidal pituitary resection for preoperative diagnosis of prolactin-secreting pituitary adenoma in women: Long term follow-up. J. Clin. Endocrinol. Metab., 81: 1711-1719.

Golden, N.H. and J.L. Carlson, 2008. The pathophysiology of amenorrhoea in the adolescent. Ann. N. Y. Acad. Sci., 1135: 163-178.

Goswami, D. and G.S. Conway, 2005. Premature ovarian failure. Hum. Reprod. Update, 11: 391-410.

Goswami, D. and G.S. Conway, 2007. Premature ovarian failure. Hormone Res., 68: 196-202.

Grinspoon, S., T. Gulick, H. Askari, M. Landt and K. Lee *et al*., 1996. Serum leptin levels in women with anorexia nervosa. J. Clin. Endocrinol. Metab., 81: 3861-3863.

Haas, V., S. Onur, T. Paul, D.O. Nutzinger and A. Bosy-Westphal *et al*., 2005. Leptin and body weight regulation in patients with anorexia nervosa before and during weight recovery. Am. J. Clin. Nutr., 81: 889-896.

Hardiman, P., O.S. Pillay and W. Atiomo, 2003. Polycystic ovary syndrome and endometrial carcinoma. Lancet, 361: 1810-1812.

Hickey, M. and A. Balen, 2003. Menstrual disorders in adolescence: Investigation and management. Hum. Reprod. Update, 5: 493-504.

Hoyt, K.L. and M.C. Schmidt, 2004. Polycystic ovary (Stein-Leventhal) syndrome: Etiology, complications and treatment. Clin. Lab. Sci., 17: 155-163.

Jones, J.M., M.L. Lawson, D. Daneman, M.P. Olmsted and G. Rodin, 2000. Eating disorders in adolescent females with and without type 1 diabetes: Cross sectional study. Br. Med. J., 320: 1563-1566.

Katzman, D.K. and N.H. Golden, 2007. Anorexia Nervosa and Bulimia Nervosa. In: Adolescent Healthcare: A Practical Guide, Neinstein, L.S. (Ed.). 5th Edn., Lippincott Williams and Wilkins, Philadelphia, PA., USA.

Kauffman, R.P. and V.D. Castracane, 2003. Premature ovarian failure associated with autoimmune polyglandular syndrome: Pathophysiological mechanisms and future fertility. J. Women's Health, 12: 513-520.

Khalid, A., 2004. Irregular or absent periods: What can an ultrasound scan tell you? Best Pract. Res. Clin. Obstet. Gynaecol., 18: 3-11.

King, J., 2006. Polycystic ovary syndrome. J. Midwifery Women's Health. 51: 415-422.

Kins, M.B., 1989. Eating disorders in a general practice population. Prevalence, characteristics and follow-up at 12 to 18 months. Psychol. Med. Monogr. Suppl., 14: 1-34.

Klingensmith, G.J., A.C. Wentz, W.J. Meyer and C.J. Migeon, 1976. Gonadotropin output in congenital adrenal hyperplasia before and after adrenal suppression. J. Clin. Endocrinol. Metab., 43: 933-936.

Koutras, D.A., 1997. Disturbances of menstruation in thyroid disease. Ann. N.Y. Acad. Sci., 816: 280-284.

Krassas, G.E., 2000. Thyroid disease and female reproduction (modern trends). Fertil. Steril., 74: 1063-1070.

Lakhani, K., A.M. Seifalain, W.U. Atiomo and P. Hardiman, 2002. Polycystic ovaries. Br. J. Radiol., 75: 9-16.

Loucks, A.B., M. Verdun and E.M. Heath, 1998. Low energy availability, not stress of exercise, alters LH pulsatility in exercising women. J. Applied Physiol., 84: 37-46.

Luciano, A.A., 1999. Clinical presentation of hyperprolactinemia. J. Reprod. Med., 44: 1085-1090.

Magiakou, M.A., G. Mastorakos, E. Webster and G.P. Chrousos, 1997. The hypothalamic-pituitary-adrenal axis and the female reproductive system. Ann. N. Y. Acad. Sci., 816: 42-56.

Marshall, L.A., 1994. Clinical evaluation of amenorrhea in active and athletic women. Clin. Sports Med., 13: 371-387.

Master-Hunter, T. and D.L. Heiman, 2006. Amenorrhea: Evaluation and treatment. Am. Fam Phys., 73: 1374-1382.

Meczekalski, B., A. Podfigurna-Stopa, A. Warenik-Szymankiewicz and A.R. Genazzani, 2008. Functional hypothalamic amenorrhoea: Current view on neuro-endocrine aberrations. Gynaecol. Endocrinol., 24: 4-11.

Misva, M., K.K. Miller, C.C. Almazan, K. Ramaswamy and A. Aggarwal *et al*., 2004. Hormonal and body composition predictors of soluble leptin receptor, leptin and free leptin index in adolescent girls with anorexia nervosa and controls and relation to insulin sensitivity. J. Clin. Endocrinol. Met., 89: 3486-3495.

Moran, C., C. Azziz, E. Carmina, D. Dewailly and F. Fruzzetti *et al*., 2000. 21-Hydroxylase-deficient nonclassic adrenal hyperplasia is a progressive disorder: A multicenter study. Am. J. Obstet. Gynecol., 183: 1468-1474.

Morrison, E., 1998. Analyzing amenorrhea. Hosp. Pract., 33: 89-100.

Oats, J. and S. Abraham, 2005. Disorders of Menstruation. In: Llewellyn-Jones Fundamentals of Obstetrics and Gynaecology, Oats, J. and S. Abraham (Eds.). Elsevier Mosby, USA., pp: 223-233.

Parl, F.F., V.E. Cruz, C.A. Cobb, C.A. Bradley and S.L. Aleshire, 1986. Late recurrence of surgically removed prolactinomas. Cancer, 57: 2422-2426.

Petraglia, F., C. Musacchio, S. Luisi and V. De Leo, 2008. Hormone-dependent gynaecological disorders: A pathophysiological perspective for appropriate treatment. Best Pract. Res. Clin. Obstetr. Gynaecol., 22: 235-249.

Popat, V.B., T. Prodanov, K.A. Calis and L.M. Nelson, 2008. The menstrual cycle: A biological marker of general health in adolescents. Ann. N. Y. Acad. Sci., 1135: 43-51.

Raber, W., A. Gessl, P. Nowotny and H. Vierhapper, 2003. Hyperprolactinaemia in hypothyroidism: Clinical significance and impact of TSH normalization. Clin. Endocrinol., 58: 185-191.

Rajkowha, M., M.R. Glass, A.J. Rutherford, K. Michelmore and A.H. Balen, 2000. Polycystic ovary syndrome: A risk factor for cardiovascular disease? Br. J. Obstet. Gynecol. Int. J. Obstetr. Gynaecol., 107: 11-18.

Rattanachaiyanont, M., S. Kunathikom, S. Angsuwattana, K. Techatraisak, O. Mekmahan, C. Karavagul and S. Ko-Anantakul, 1997. Primary amenorrhoea: A retrospective study at Siriraj hospital. J. Med. Assoc. Thai, 80: 619-625.

Reinhold, C., H. Hricak, R. Forstner, S.M. Ascher, P.M. Bret, W.R. Meyer and R.C. Semelka, 1997. Primary amenorrhea: Evaluation with MR imaging. Radiology, 203: 383-390.

Rosmark, B., C. Berne, S. Holmgren, C. Lago, G. Renholm and S. Sohlberg, 1986. Eating disorders in patients with insulin-dependent diabetes mellitus. J. Clin. Psychiatry, 47: 547-550.

Rothman, M.S. and M.E. Wierman, 2008. Female hypogonadism: Evaluation of the hypothalamic-pituitary-ovarian axis. Pituitary, 11: 163-169.

Rydall, A.C., G.M. Rodin, M.P. Olmsted, R.G. Devenyi and D. Daneman, 1997. Disordered eating behavior and microvascular complications in young women with insulin-dependent diabetes mellitus. N. Engl. J. Med., 336: 1849-1854.

Schlechte, J.A., B.M. Sherman, F.K. Chapler and J. Van Gilder, 1986. Long term follow-up of women with surgically treated prolactin-secreting pituitary tumors. J. Clin. Endocrinol. Metab., 62: 1296-1301.

Schrager, S. and L. Sabo, 2001. Sheehan syndrome: A rare complication of postpartum hemorrhage. J. Am. Board Fam Pract., 14: 389-391.

Snyder, P.L., B.F. Fowbe, N.I. Schatz, P.J. Savino and T.A. Gennarelli, 1986. Hypopituitarism following radiation therapy of pituitary adenomas. Am. J. Med., 81: 457-462.

Speroff, L. and M.A. Fritz, 2005. Amenorrhea. In: Clinical Gynecologic Endocrinology and Infertility, Speroff, L. and M.A. Fritz (Eds.). 7th Edn., Lippincott Williams and Wilkins, Philadelphia, PA., USA., pp: 401-464.

Speroff, L., R.H. Glass and N. Hase, 1999. Hypothalamic Amenorrhoea. In: Clinical Gynecologic Endocrinology and Infertility, Speroff, L., R.H. Glass and N. Hase (Eds.). Lipponcott Williams and Wilkins, USA.

Spesier, P.W. and P.C. White, 2003. Congenital adrenal hyperplasia. N. Engl. J. Med., 349: 776-788.

Van Kasteren, Y.M., R.D. Hundscheid, A.P. Smits, F.P. Cremers, P. van Zonneveld and D.D. Braat, 1999. Familial idiopathic premature ovarian failure: An overrated and underestimated genetic disease? Hum. Reprod., 14: 2455-2459.

Venturoli, S., E. Porcu, R. Fabbri, R. Paradisi and S. Ruggeri *et al*., 1987. Menstrual irregularities in adolescents: Hormonal pattern and ovarian morphology. Horm. Res., 24: 269-279.

Welt, C.K., J.L. Chan, J. Bullen, R. Murphy and P. Smith *et al*., 2004. Recombinant human leptin in women with hypothalamic amenorrhea. N. Engl. J. Med., 351: 987-997.

Wieck, A. and P.M. Haddad, 2003. Antipsychotic-induced hyperprolactinaemia in women: Pathophysiology, severity and consequences. Selective literature review. Br. J. Psychiatry, 182: 199-204.

Wilson, G.R., J.E. Haddad and C.J. Haddad, 2005. Amenorrhea: Common causes and evaluation. Compr. Ther., 31: 270-278.

Yen, S.S., 1998. Effects of lifestyle and body composition on the ovary. Endocrinol. Metab. Clin. North Am., 27: 915-926.

Zargar, A.H., B.A. Laway, S.R. Masoodi, M. Salahuddin and M.A. Ganie *et al*., 2004. Clinical and endocrine aspects of pituitary function. Saudi Med. J., 24: 1428-1432.