East African Medical Journal Vol. 94 No. 7 July 2017
PATHOGENESIS OF OVARIAN CANCER: CURRENT PERSPECTIVES
Jacqueline J. Chesang, MBChB, MMED (OBS/GYN), PhD, P.O Box 25636 Nairobi 00603

PATHOGENESIS OF OVARIAN CANCER: CURRENT PERSPECTIVES

J.J. Chesang

ABSTRACT

Objective: **To present a review of current knowledge of the pathogenesis of ovarian cancer and its clinical implications.**
Data Source: **Extensive literature search was conducted to identify relevant studies.**
Study Selection: Studies in the English language about or related to pathogenesis of ovarian cancer were selected.
Data Extraction: **Applicable findings from selected studies were extracted as reported in the studies.**
Data Synthesis: **Proposed theories of pathogenesis, and source population of ovarian tumour cells are discussed, and classification of ovarian cancer presented. Clinical implications of these new insights are also presented. Traditionally, epithelial ovarian tumours are thought to arise from the ovarian surface epithelium. Proposed factors that drive ovarian tumourigenesis include incessant-ovulation, gonadotropin stimulation, inflammation, and hormonal stimulation. There is evidence suggesting that ovarian tumour cells arise from extra-ovarian sites with secondary involvement of the ovary; the most prevalent and aggressive histological type (serous tumours) being of tubal origin. Ovarian cancer is now classified as type I and II or type I, II and III on the basis of shared mutations, histogenetic pathways, and behaviour.**
Conclusion: **The pathogenesis of ovarian cancer has not been established. Adoption of extra-ovarian origin of tumour cells may have implications for prevention, screening, and treatment of ovarian cancer.**

Key Words: Ovarian cancer, pathogenesis, Müllerian, extra- Müllerian, extra-ovarian, incessant-ovulation, ovarian surface epithelium, hormonal.

INTRODUCTION

Worldwide, over 225,000 women are diagnosed with, and over 140,000 women die from ovarian cancer each year [1, 2]. Despite advances in treatment, ovarian cancer causes considerable morbidity, and long-term survival remains poor. The primary reasons are lack of early-detection methods [3] (most ovarian tumours [75%] are diagnosed at an advanced stage [stage III and IV] [4]) and lack of an understanding of its pathobiology [5]. Currently, there is ongoing debate regarding ovarian cancer pathogenesis [6]. Understanding the pathogenesis of a disease is important in developing effective screening, preventive and therapeutic approaches. Herein,

possible pathogenetic pathways, source of ovarian tumour cells, change in classification of epithelial ovarian tumours, and their implications on prevention, screening, treatment and future research directions are discussed.

OVARIAN SURFACE EPITHELIUM AS A SOURCE OF OVARIAN TUMOUR CELLS

Epithelial ovarian tumours have been thought to arise from the ovarian surface epithelium (OSE), which is of coelomic origin. In this tumourigenic pathway, the OSE invaginates forming cortical inclusion cysts (CICs); then, under the influence of local factors (possibly steroidal hormones), it undergoes metaplasia (coelomic metaplasia theory) followed by neoplastic transformation [7]. Theories have been put forward to explain this.

INCESSANT-OVULATION THEORY

Incessant-ovulation is thought to play a role in ovarian carcinogenesis. Ovulation includes inflammatory processes, with leukocyte infiltration, and release of inflammatory mediators [8]. Ovarian surface epithelial (OSE) cells adjacent to the site of ovulation are exposed to inflammatory mediators and reactive oxidants that can cause DNA damage [9, 10]. One source of possibly damaging free radicals is thought to be leukocytes that infiltrate developing follicles [9]. Inflammatory processes have been implicated in the pathogenesis of ovarian cancer [11], which is supported by the observed lower risk associated with prolonged use of anti-inflammatory drugs (i.e., NSAIDS, including ASA) [10]. Conversely, asbestos and talc (which induce inflammatory responses) are thought to increase the risk of ovarian cancer [10] although the relationship between talc and ovarian cancer is inconclusive [12].

Following ovulation, epithelial cells replicate to cover the defect on the ovarian surface [9, 13]. The higher proliferative rate decreases the chance of DNA repair or apoptosis of cells that have sustained genomic damage [14]. In addition, formation of the corpus luteum is also associated with angiogenesis [8]. In the incessant-ovulation theory, the greater the total number of life-time ovulatory cycles, the higher the risk. Lifetime duration of ovulation has been shown to be associated with the risk of ovarian cancer at a mean of 6% per year, with the 20-29 year age group being at the highest risk, namely 20% per year of ovulation [15].

It has been suggested that ovarian cancer arises from CICs, [16, 17] and this is supported by the observation that early ovarian cancer is confined to the organ without surface involvement [18]. CICs are thought to form during the ovulatory-repair process [16, 17]. In support of this is a study in which the number of CICs was positively associated with ovulatory age: 25-34 year-old women had a mean of 2.6 CICs, whereas those of 55 years and above had a mean of 4.5 CICs (Spearman's rho = 0.2; P = 0.06). There were also fewer CICs in ever-users of oral contraceptives. However, this study lacked power to assess weak associations and did not include young women [17]. Extra-ovarian sources of CICs have also been proposed [18] (this will be discussed later).

Consistent with the incessant-ovulation hypothesis are the plausible protective effects of oral contraceptives (OCs) use, breastfeeding, and parity, conditions in which ovulation is suppressed [9, 10, 14, 18]. However, several factors serve to undermine the incessant-ovulation theory. Most importantly, the strengths of the inverse association between OCs use and parity and ovarian cancer far outweigh that which can be attributed to inhibition of ovulation alone [8, 18-20]. Women ovulate

for an average of 20 years in their lifetime. Each full-term pregnancy inhibits ovulation for about 1 year, which reduces lifetime ovulations by 5-6%. It has been shown that, in parous women, for each additional pregnancy, the reduction of risk of ovarian cancer is 14-16%. Similarly, OCs are inversely associated with risk at 9% per year of use, which is greater than the 5-6% protection expected from inhibition of ovulation [16].

Secondly, women with polycystic ovary syndrome (PCOS), a condition characterised by decreased ovulation or anovulatory cycles, have an increased risk of ovarian cancer [8, 10, 14, 16, 21]. Inhibition of ovulation alone also does not explain the more marked inverse association with twin pregnancies, given that twin pregnancies are associated with increased ovulation [18], nor the inverse association with tubal ligation and hysterectomy [21]. Furthermore, normal and malignant OSE cells have receptors for follicle stimulating hormone (FSH), luteinising hormone (LH), gonadotrophin releasing hormone(GnRH), oestrogens, progesterone, and androgens, suggesting a role for these hormones in ovarian carcinogenesis [12, 22].

GONADOTROPIN HYPOTHESIS

Excessive gonadotropin levels are thought to play a facilitating role in the development of ovarian cancer [14, 21]. In this case, gonadotropins, acting directly or via increased production of oestrogen, stimulate malignant transformation of ovarian epithelial cells [8, 14, 16, 18]. It is also plausible that gonadotropins, by stimulating ovulation could indirectly promote ovarian cancer development [23].

Consistent with the gonadotropin hypothesis are: the protective effects of OCs and pregnancy, both of which decrease the levels of gonadotropins [14, 16, 23]; the higher risk with infertility treatment, which is associated with increased gonadotropin levels [13, 24]; the higher risk in PCOS, a condition associated with elevated LH levels [8, 12, 14, 18]; and the observation that the incidence of ovarian cancer peaks 10-20 years after menopause, when gonadotropin levels are elevated [24].

The gonadotropin theory has its shortcomings. Firstly, post-menopausal hormone (PMH) is associated with higher ovarian cancer risk despite decreasing gonadotropin levels [13, 14, 18, 22]. Secondly, the gonadotropin theory does not explain the inverse association of twin pregnancies with ovarian cancer, which are usually associated with increased gonadotropin levels [18]. Thirdly, there is an inverse association with breastfeeding although it is associated with elevated levels of FSH [8].

HORMONE-STIMULATION HYPOTHESIS

Hormones are thought to increase the risk of cancer, at least in part, by increasing the proliferation rate of cells in the organs under their influence. This increases the probability of occurrence of genetic errors that lead to neoplasia and of the affected cells undergoing replication before DNA repair [19, 24].The hormone-stimulation hypothesis suggests that excessive oestrogen or androgen stimulation of the OSE promotes neoplastic transformation, whereas progesterone is protective.

Oestrogens

Oestrogens have been shown to enhance proliferation of normal and malignant human OSE cells [21, 24]. It has also been proposed that oestrogen may promote ovarian carcinogenesis by inhibiting apoptosis of ovarian epithelial cells. This was demonstrated in a study done on macaques (*Macacafascicularis*), in which those treated with oestradiol alone or Triphasil (Triphasil contains ethinyloestradiol and levonorgestrel) had

lower rates of apoptosis, 1.8% and 14.5% respectively, than the group treated with levonorgestrel alone, 24.9% [20].

Oestrogen is thought to play a promotional role in ovarian carcinogenesis. The observed inverse association with breastfeeding is consistent with this. In breastfeeding, oestradiol and LH levels are decreased, but FSH is elevated. In addition, use of PMH is associated with higher risk of ovarian cancer and the risk may be higher for those who used oestrogen-alone, as opposed to oestrogen-progesterone therapy [25, 26]. Oestrogen replacement therapy decreases gonadotropin levels but raises circulating oestradiol levels [16].

It has been argued that it is the hormone levels in the ovarian microenvironment, as opposed to circulating hormone levels, that influence ovarian cancer risk (stromal hypothesis) [27]. Levels of oestrogen in the ovarian stroma are ≥100-fold higher than that in the circulation, with follicular levels being markedly higher still [27]. At the time of ovulation, OSE cells are exposed to follicular fluid that contains concentrations of oestradiol about 10,000 times higher than serum levels [16]. It has been postulated that DNA damage of OSE cells at the ovulation site/CICs may be caused by the exposure to high oestrogen levels [27]. It has also been shown that ovarian cancer cells produce oestrogen, further supporting a paracrine role of this hormone in the development of ovarian cancer [27]. Pregnancy is established as protective against ovarian cancer. During pregnancy, however, the circulating oestrogen levels are about 100-fold higher than the non-pregnant levels, which seems quite inconsistent with a promotional role for oestrogen in ovarian cancer; however, after the first few weeks of pregnancy, hormone production, including oestrogen, is mainly from the placenta [18]. According to the stromal hypothesis, exposure of CICs to high oestrogen levels may lead to neoplastic transformation [14]. Congruent with this, it has been argued that the protection conferred by combined OCs via decreased ovulation can also be attributed to reduced levels of oestrogen within ovarian tissue [27].

Androgens

High androgen levels are thought to play a facilitating role in ovarian cancer development [14, 16]. Androgens have been shown to promote proliferation of normal OSE cells [18]. High levels of androgens are produced by developing follicles [10]. Androgen is the hormone with the highest concentration in the follicular fluid, being 10 times higher than that of oestradiol in early follicles (<10mm) [16]. Therefore, CICs are exposed to an androgen-rich microenvironment during the follicular phase [10, 16]. Furthermore, follicles that later undergo atresia continue to produce androgens [16]. These observations lend further support to the stromal hypothesis [18].

In support of the role of androgens increasing ovarian cancer risk, there is a higher risk of ovarian cancer in women with PCOS, hirsutism, and acne, conditions associated with elevated androgen levels [8, 10, 16]. In PCOS, the LH and androgen levels are raised, whereas FSH levels are low [28]. The increased risk of ovarian cancer in post-menopausal or obese women has also been linked to increased androgen levels [14]. During menopause, the raised gonadotropin levels stimulate production of androgens from the ovary, resulting in an androgen-rich microenvironment [18, 24]. Use of OCs results in a decrease in ovarian testosterone production and this is thought to contribute to its protective effect [8, 16, 18].

Progesterone

Progesterone has been shown to have a protective role in ovarian carcinogenesis [16, 20]. The protective effect of progesterone is thought to be mediated via decreased

proliferation and increased apoptosis [27]. By increasing apoptosis of ovarian epithelial cells, progestins may increase the likelihood that cells that have sustained DNA damage will be eliminated, thus decreasing the risk of ovarian cancer [20].

The following observations are compatible with the proposition that progesterone is protective. First, the risk of ovarian cancer is inversely related to parity [12]. The inverse association with pregnancy surpasses that which can be attributed to ovulation inhibition and has been attributed to the high levels of progesterone in pregnancy [16]. Second, compared to singleton pregnancies, the reduction in risk of ovarian cancer may be higher in women with dizygotic twins [16, 18, 27], with one study reporting a parity-adjusted OR of 0.74 (95% CI = 0.55 – 1.01) [29]. This is despite these women having higher gonadotrophin levels and being more likely to double-ovulate during their reproductive years than women with singleton pregnancies only. This protection has been attributed to higher progesterone levels in twin pregnancies [16, 18, 27]. The high levels of progesterone are thought to cause exfoliation of cells thereby reducing the ovarian burden of cells with genetic damage [24]. Consistent with this, studies have reported lower risk of ovarian cancer with older age at first and last birth [30-32]. Third, use of PMH is associated with higher risk of ovarian cancer and the risk may be higher for those who used oestrogen alone as opposed to oestrogen/progesterone combinations [25, 26]. It has also been suggested that the progestin component of OCs importantly contributes to lower risk of ovarian cancer [16, 20].

Current evidence suggests a promoting role for high levels of androgens, oestrogens, and gonadotropins, whereas ovulation inhibition and high progesterone levels are inversely associated. However, it is difficult to tease apart the effects of the different hormones on risk of ovarian cancer because they are closely interrelated [18]. These hormones have a common synthetic pathway. In addition, circulating levels of one hormone may influence the production of another, by exerting negative or positive feedback on the hypothalamo-pituitary-ovarian axis. For example, androgens are a precursor of oestrogens, high levels of oestrogen result in inhibition of gonadotropin production, whereas high gonadotropin levels drive the production of oestrogens and androgens. In addition, ovulation, which is thought to play an important role in ovarian cancer causation, affects the levels of various hormones. In pre-menopausal women, anovulation results in high levels of androgens and oestrogens and low levels of progesterone. Furthermore, established risk factors support more than one hypothesis of ovarian cancer pathogenesis. Finally, steroid-hormone receptors, although they preferentially bind to specific ligands are, nonetheless, somewhat promiscuous and can bind and transmit signals from more than one hormone.

Most epithelial ovarian cancers resemble epithelial cells found in other sites: serous tumours resemble the fallopian tube epithelium; mucinous tumours resemble endocervical or intestinal epithelia; endometrioid tumours resemble the endometrial lining; and transitional cell tumours resemble the urothelium [4, 33-35]. To explain this, it has been argued that the OSE first undergoes metaplasia into Müllerian type cells, followed by neoplastic transformation (the coelomic metaplasia theory) [7].

EXTRA-OVARIAN ORIGIN OF OVARIAN TUMOUR CELLS

The fact that ovarian tumours do not bear resemblance to cells normally found in the ovary, and the fact that no precursor lesions have been identified within the ovary for

type II tumours have led to questions regarding the origin of cancer cells [36, 37]. It has been postulated that epithelial ovarian tumours arise from extra-ovarian sites with secondary involvement of the ovary.

MÜLLERIAN ORIGIN THEORY

In the Müllerian-origin theory, pelvic serous carcinomas are thought to arise from the fallopian tube (tubal-origin theory). Pelvic or extra-uterine serous carcinoma is classified as serous ovarian carcinoma, fallopian tube serous carcinoma (FTSC), and primary peritoneal serous carcinoma based on the presumed site of origin. The criteria used for the classification of pelvic serous carcinomas are biased towards an over-diagnosis of ovarian serous carcinoma at the expense of tubal and peritoneal [37-40]. The tubal-origin theory is based on histological findings of early lesions, known as serous tubal intraepithelial carcinomas (STICs), in the fallopian tubes of women with *BRCA1* or *BRCA2* mutations undergoing prophylactic surgery (bilateral salpingo-oophorectomy). STICs, mainly located in the distal end of the fallopian tube (fimbria), arise from secretory cells of the tubal epithelium (tubal epithelium has secretory cells and ciliated cells), are positive for *p53*, and are considered precursor lesions to serous carcinoma [37, 39, 40]. STICs are also associated with pelvic serous carcinoma in patients with unknown *BRCA* mutation status [36].

STICs have also been found in association with ovarian and primary peritoneal high-grade serous carcinoma (HGSC). In one study, 37% of cases with primary peritoneal serous cancer had STICs. This is in spite of the study being limited by variation in the degree of analysis of the fallopian tube (it is expected that extensive examination of the fallopian tube would have yielded a higher number of STICs). In addition, those with STICs were significantly older (P = 0.007) and more likely to have stage IV disease (P = 0.037) than those without STICs [40]. Age is a risk factor for epithelial ovarian cancer and high-grade serous ovarian carcinoma (HGSOC) is usually diagnosed at an advanced stage. In studies where the fallopian tubes were comprehensively examined, using the Sectioning and Extensively Examining the Fimbriae (SEE-FIM) protocol (SEE-FIM protocol increases the surface area of the fimbria under examination by 60%) [41], STICs were found in 50 to 60% of women with ovarian and peritoneal high-grade serous carcinoma without known genetic predisposition to ovarian cancer [37]. No similar lesions have been found in the ovaries and no precursor lesions have been found in studies of contralateral ovaries of women with non-familial unilateral ovarian cancer [7]. Therefore, pelvic serous carcinoma is thought to arise from the fallopian tube, the ovary being a secondary site.

STICs have been found in direct continuity with short stretches of secretory cells with positive nuclear staining for p53, termed "p53 signatures" [36]. p53 signatures have also been found in the absence of STICs or cancer [7].*p53* mutations have been reported in 57% of cases of p53 signatures [42]. *p53* is a tumour suppressor gene located on chromosome 17. Its protein product, tumour protein 53 (TP53 also known as p53), halts the cell cycle to allow for repair of damaged DNA, or induces apoptosis. Its cellular levels rise in response to DNA damage [43]. *p53* mutations are associated with many human cancers [16, 28, 43]. p53 signatures are more common in the fallopian tube epithelium as demonstrated by a study in which, out of 75 *BRCA* mutation carriers examined, 29 p53 signatures were found in the fallopian tubes, 1 in the OSE, and none in CICs [44].

The tubal-origin theory proposes that pelvic serous carcinomas arise from the

distal tubal epithelium. The finding of p53 signatures in continuity with STICs has led to the conclusion that p53 signatures precede STICs [37]. With respect to the natural history of cancer, an ovarian carcinogenesis model has been proposed, starting from DNA damage, followed by p53 signatures which progress to STICs, and finally to invasive cancer [7, 39]. Support for the link between STICs, p53 signatures, and invasive tubal carcinoma is by the demonstration that they arise from secretory cells, are mainly found on the tubal fimbriae, and when concurrent, express similar *p53* mutations [39, 40, 45]. In addition, STICs, HGSOC, primary peritoneal serous carcinoma, and FTSC have similar *p53* mutations in some cases [39]. Furthermore, the peritoneum is lined by mesothelium and therefore, malignant mesothelioma and not serous carcinoma is expected [40]. If serous carcinoma is of tubal origin, this would explain the advanced stage at diagnosis because the cancer cells, arising from the tube, have access to the entire peritoneal cavity [7, 40].

The prevalence of p53 signatures in non-neoplastic fimbria has been found to be the same in women with and without *BRCA* mutation, one third in each [44]. This suggests that factors influencing p53 signatures are independent of *BRCA* mutation status. It is plausible that p53 signatures are a result of a physiological increase in TP53, in response to DNA damage [7, 45]. The observed higher risk of ovarian cancer in *BRCA* mutations carriers suggests that *BRCA* mutations influence progression of p53 signatures to STICs, and eventually invasive cancer [36].

Clear-cell carcinoma and endometrioid tumours have been associated with implants of endometriosis elsewhere in the pelvis and are, therefore, thought to arise from the endometrium [7, 37]. A possible cause of endometriosis, though not established, is thought to be retrograde menstruation [46]. Supporting the association with endometriosis is the observation that tubal ligation decreases the risk of clear-cell and endometrioid carcinoma but no other epithelial types [37]. In a nationwide population-based study in Australia, overall, there was a statistically non-significant association (OR = 1.31; 95% CI = 0.97-1.78). However, a statistically significant higher risk of endometrioid (OR = 1.85; 95% CI = 1.02-3.38) and clear-cell carcinoma was observed (OR = 2.66; 95% CI = 1.31-5.44) [47]. In the Müllerian-origin theory, mucinous tumours are thought to arise from the endocervix. The Müllerian-origin theory is supported by the observed protection conferred by tubal ligation and hysterectomy, without oophorectomy [37]. This protection has also been attributed to blockage of carcinogens, from the external environment, from reaching the ovaries via the Müllerian tract [48].

The Müllerian-origin theory is limited by the fact that women with *BRCA1* or *BRCA2* mutation who undergo prophylactic surgery are still at risk of peritoneal serous carcinoma [38]. Furthermore, mucinous tumours resemble the gastrointestinal mucosa and the origin of Brenner tumours, which resemble the urothelium, has not been explained [7, 37]. These findings suggest that there could still be other sources of cancer cells.

SECONDARY MÜLLERIAN-ORIGIN THEORY

The secondary Müllerian-origin theory proposes that ovarian cancer arises from vestigial embryological remnants of the Müllerian system, which can act as a source for all the epithelial cell types of ovarian cancer. Microscopic vestigial embryological remnants lined by Müllerian epithelial cells (including endosalpingiosis, endocervicosis, and endometriosis), which are collectively referred to as the "secondary Müllerian

system," are found in the paraovarian and paratubal areas and also within the ovarian cortex and medulla [38]. Brenner and mucinous tumours are associated with, and thought to arise from, nests of transitional-type epithelial cells: Walthard cell-nests, located at the tubal-peritoneal junction (paraovarian and paratubal areas), and hence, their non-Müllerian appearance. Previously, mucinous tumours had been thought to originate from the endocervix but, due to their non-Müllerian appearance the cell-nest theory is preferred. Mucinous tumours more closely resemble gastrointestinal mucosa than endocervix [7, 37]. Since the secondary Müllerian system is also found within the ovary, albeit rarely, ovarian cancer can be viewed as originating from the ovary in this instance [38].

The theory of extra-ovarian origin of epithelial ovarian tumours is limited by the observed high levels of ovarian involvement. Although serous tumours are thought to arise from the tubal epithelium, serous tumours involve the ovaries and other pelvic organs more extensively than the tubes. In addition, endometrioid and clear-cell tumours are usually confined to the ovaries although endometriosis usually involves multiple sites in the pelvis. It has also been suggested that, rarely, low-grade serous cancer (LGSC) progresses to HGSC [37]. This is due to the finding of HGSCs in association with serous borderline tumours and LGSCs, with identical *KRAS* mutations and no *p53* mutations [7].

RECONCILING INCESSANT-OVULATION, HORMONAL, AND EXTRA-OVARIAN ORIGIN THEORIES

The most prevalent and highly fatal, serous type of ovarian cancer, is now postulated to be of tubal origin [40]. A relevant question is therefore: what is the effect of ovulation on the tubal cells?

The tubal fimbriae lie in close spatial proximity to the ovaries and may, therefore, be exposed to substances produced during ovulation [49]. In a study of mice and baboons, ovulation was found to increase both macrophage infiltration into the fallopian tube, and DNA damage of tubal epithelial cells (TEC). Macrophages produce substances that promote inflammation. No increase in proliferation of TEC was noted. However, these effects were assessed at 12 hours and 16 hours after injection of hCG which does not rule out the possibility of proliferation occurring later in the post-ovulatory period. In addition, exposure of cells to hydrogen peroxide led to an increase in DNA damage [34]. Hydrogen peroxide is used to mimic the oxidative stress caused by ovulation. On the other hand, menopausal levels of gonadotropins (FSH and LH), and oestradiol (10nM) did not have genotoxic or proliferative effects on TEC in spite of the presence of receptors for these hormones on these cells (FSHR, LHR, and ERα) [49].

In a study of *BRCA* mutation carriers, p53 signatures were statistically significantly associated with low parity and older age at first childbirth. Compared to nulliparous women, women with a parity of 3 or more had an OR of 0.2 (95% CI = 0.04-0.9; P-trend = 0.02). This study was limited by a small sample size and missing data. Furthermore, no association was found between p53 signatures and OC use, or age [50]. Therefore, exposure of tubal cells to substances produced during ovulation may lead to neoplastic transformation of tubal cells, which then seed to the ovary.

The coelomic metaplasia theory proposes that the OSE undergoes metaplasia under hormonal influence, with metaplasia being more likely in CICs, into Müllerian-type cells followed by neoplastic transformation [16, 38]. However, due to the intimate contact between the fallopian tubes and the ovary during ovulation, it is conceivable that dislodged normal tubal

epithelial cells can get incorporated into the disrupted OSE, forming inclusion cysts, which later undergo neoplastic transformation [37, 51]. LGSC and HGSC can then develop via different pathways [7]. In support of this theory are the findings of a study done by Li et al. in which a majority of ovarian sections examined (46 of 48 cases [96%]) were lined only by OSE of mesothelial phenotype; in contrast, 78% (667 of 856 cases) of epithelial inclusions were of fallopian tubal phenotype [51].

If ovarian cancer cells arise from extra-ovarian sites, why is the ovary the most affected organ? In a case-control study, Pearce et al. found a positive association between LGSC and a history of endometriosis (OR = 2.11; 95% CI = 1.39 - 3.20), which is unusual: endometriosis is associated with higher risk of ovarian cancer, specifically the endometrioid and clear-cell histologic subtypes. To explain this, they hypothesised that the LGSC could arise from endosalpingiosis which is usually asymptomatic, and therefore, difficult to study using a case-control design. They further postulated that the host's susceptibility to implants of endometrial tissue may also be responsible for an increased risk of endosalpingiosis, which is as a result of implantation of cells from the fallopian tube, both being of Müllerian origin [52]. Therefore, it is possible that the ovaries of women who develop ovarian cancer provide a suitable microenvironment for the development of precancerous cells [48]. Indeed, it has been shown that local microenvironment can influence the transformation of normal cells to malignant cells. Conversely, the microenvironment can also cause malignant cells to differentiate into normal cells [53]. It has also been argued that secondary sites of cancer metastasis do not occur by chance. Metastatic cancer cells develop only in sites that provide a suitable environment – the "seed and soil" hypothesis of metastatic spread [43].

CLASSIFICATION OF OVARIAN CANCER

Recent studies have led to the classification of epithelial ovarian cancer into type I and type II tumours based on shared mutations, histogenetic pathways, and behaviour [36, 37, 54]. Type I tumours include LGSC, low-grade endometrioid, Brenner tumours, clear-cell, and mucinous carcinomas. They are generally low grade, slow growing, present at an early stage (stage I), are relatively stable genetically, and have a good prognosis. They are associated with mutations in *KRAS, BRAF, ERB2, PTEN, PIK3CA, ARIDIA*, and *CTNNB1/β-catenin*, and rarely have *p53* mutations [36, 37, 39, 40]. Type I tumours have been found adjacent to benign epithelial tumours [39], and are thought to develop sequentially from CICs, to borderline tumours, and finally to invasive tumours [36, 37, 40]. Type II tumours are comprised mainly of HGSOC, others are high-grade endometrioid, malignant mixed mesodermal tumours (carcinosarcomas), and undifferentiated carcinomas [37]. They present in advanced stage (stage II-IV), grow rapidly, are genetically unstable, and show *p53* mutations. They rarely harbour mutations found in type I tumours and are not found with adjacent borderline tumours [36, 37]. Type I tumours account for 25% of all epithelial ovarian cancers while type II account for the other 75% [55]. Ovarian tumours can also be classified as type I (low-grade serous and mucinous tumours), type II (endometrioid and clear-cell tumours), and type III (high-grade serous and undifferentiated tumours) [56, 57]. Type III tumours have the worst prognosis [57].

CLINICAL IMPLICATIONS

In view of current evidence supporting the protective role of progesterone, this hormone could be used in prevention. It has been proposed that high levels of progesterone, equivalent to those found in pregnancy, could be administered periodically to women at high risk of ovarian cancer so as to trigger apoptosis of OSE and therefore, promote loss of cells that may have undergone genetic damage [27]. If used at all, it may also be safer to use progesterone in combination with oestrogen as PMH [24]. Progesterone could also be used in combination with other cytotoxics in the treatment of ovarian cancer [27].

Tamoxifen (a second generation anti-oestrogen) has been used as a second-line treatment in patients with ovarian cancer with little success (5-18% response rates) [3, 13, 27]. These poor results could be attributed to choice of patients in clinical studies; tamoxifen is used as a treatment of last resort [3, 13]. As response to treatment is related to the level of ER, another factor could be low levels of ER in ovarian cancer cells [13, 27]. It is also not clear whether tamoxifen acts as a pure antagonist of oestrogen on ovarian tissue or if it also has an agonist effect [3]. Tamoxifen used in combination with platinum-based chemotherapy has shown a better response (overall response of 50%) [27]. GnRH analogues, progestins, anti-androgens, and letrozole have also been used in treatment of recurrent ovarian cancer with poor results [12]. Letrozole, an aromatase inhibitor, has been shown to increase the levels of ER in ovarian tumours which may promote the response to anti-oestrogens [27]. It has also been proposed that oestrogen-regulated gene products including fibulin-1 and cathepsin D, and ERβ-negative status could be used as early biomarkers of ovarian cancer [3].

With the Müllerian- and secondary Müllerian-origin theories, germ-cell and gonadal stromal tumours are the only tumours thought to be of true primary ovarian origin [37]. Both theories suggest that both type I and type II tumours arise from extra-ovarian sites with secondary involvement of the ovary. Adoption of this concept will have implications for prevention, screening, and treatment of ovarian cancer. It may also influence nomenclature of ovarian tumours, and impact on future research directions.

Prophylactic surgery for women with familial predisposition to ovarian cancer may be limited to salpingectomy or fimbriectomy[37-40], sparing the ovaries, and therefore, preserving fertility (this can be achieved through assisted reproductive technology) and avoiding early menopause with its attendant risks such as osteoporosis, vasomotor instability, and an increased incidence of cardiovascular diseases [28]. In addition, if the tumour arises from the tubal fimbria, fimbriectomy could be an option for patients who desire tubal ligation as a method of contraception, while warning on the irreversibility of the procedure, as this would further decrease the risk of ovarian cancer [39]. Knowledge of the pathogenesis of ovarian cancer will also help in understanding the mechanism of the risk factors and, therefore, aid in the development of effective preventive strategies [38].

Based on the view that epithelial ovarian cancer arises from OSE, screening has focused on early detection of ovarian cancer while it is still confined to the ovary. With appreciation of the dualistic model of pathogenesis, one screening test may not detect all the different types of ovarian cancer. Type I tumours, which account for 25% of ovarian cancer and 10% of deaths, are slow growing, and can be detected at an early stage by pelvic examination, and transvaginal ultrasound. Therefore,

development of a screening biomarker is not urgent for this group of tumours. Future research on screening should be more focussed on type II tumours, which are rapidly progressing and account for 75% of the disease burden and 90% of deaths from ovarian cancer. The aim of screening should be to detect the tumour at low-volumes, rather than at an early stage, by developing sensitive and specific biomarkers that are expressed early in ovarian carcinogenesis [37]. It is also important to determine the time interval between the development of STICs and peritoneal spread and to develop tests with high sensitivity and specificity for detecting STICs [36].

In addition, different therapeutic approaches should be used for type I and type II tumours. Surgery is generally effective for type I tumours as they are usually detected at an early stage. At an advanced stage, type I tumours, due to their slow growing nature, may not be responsive to the chemotherapeutic agents that are effective for the rapidly proliferating type II tumours [37]. A different therapeutic approach is, therefore, needed for advanced type I tumours. Surgery in type II tumours, HGSOC, is usually limited to debulking due to the fact that they are usually detected at an advanced stage.

Staging of ovarian cancer may also be affected, with proposals of staging according to tumour bulk [7]. In addition, various names have been suggested in place of ovarian cancer, including "extra-uterine Müllerian carcinoma", to reflect the origin of the tumour cells. Pelvic serous carcinomas arise from the fallopian tube (tubal-origin theory) and should therefore be treated as one disease entity [38].

CONCLUSION

All said, it is important to note that the true pathogenesis of ovarian cancer has not been established. Understanding the pathogenesis of a disease is important in the development of effective screening and therapeutic methods, as well as preventive strategies. Currently, there is no effective screening method for ovarian cancer and current therapeutic approaches have had little success.

REFERENCES

1. Jemal, A., M.M. Centre, C. DeSantis, and E.M. Ward, *Global patterns of cancer incidence and mortality rates and trends.* Cancer Epidemiology, Biomarkers and Prevention, 2010. 19(8): p. 1893-907.
2. Jemal, A., F. Bray, M.M. Center, J. Ferlay, E. Ward, and D. Forman, *Global cancer statistics.* CA: A Cancer Journal for Clinicians, 2011. 61(2): p. 69-90.
3. Cunat, S., P. Hoffmann, and P. Pujol, *Estrogens and epithelial ovarian cancer.* Gynecologic Oncology, 2004. 94(1): p. 25-32.
4. Jelovac, D. and D.K. Armstrong, *Recent progress in the diagnosis and treatment of ovarian cancer.* CA Cancer Journal for Clinicians, 2011. 61(3): p. 183-203.
5. Ness, R.B., M.T. Goodman, C. Shen, and R.C. Brunham, *Serologic evidence of past infection with Chlamydia trachomatis, in relation to ovarian cancer.* The Journal of Infectious Diseases, 2003. 187(7): p. 1147.
6. Rosner, B.A., G.A. Colditz, P.M. Webb, and S.E. Hankinson, *Mathematical models of ovarian cancer incidence.* Epidemiology, 2005. 16(4): p. 508-15.
7. Kurman, R.J. and I.M. Shih, *The origin and pathogenesis of epithelial ovarian cancer: a proposed unifying theory.* American Journal of Surgical Pathology, 2010. 34(3): p. 433-443.
8. Fleming, J.S., C.R. Beaugié, I. Haviv, G. Chenevix-Trench, and O.L. Tan, *Incessant ovulation, inflammation and epithelial ovarian carcinogenesis: revisiting old hypotheses.* Molecular and Cellular Endocrinology, 2006. 247(1–2): p. 4-21.
9. Murdoch, W.J., *Carcinogenic potential of ovulatory genotoxicity.* Biology of Reproduction, 2005. 73(4): p. 586-590.
10. Saad, A.F., W. Hu, and A.K. Sood, *Microenvironment and pathogenesis of epithelial*

ovarian cancer. Hormones and Cancer, 2010. 1(6): p. 277-290.
11. Akhmedkhanov, A., P. Toniolo, A. Zeleniuch-Jacquotte, I. Kato, K.L. Koenig, and R.E. Shore, *Aspirin and epithelial ovarian cancer.* Preventive Medicine, 2001. 33(6): p. 682-7.
12. Gadducci, A., S. Cosio, A. Gargini, and A.R. Genazzani, *Sex-steroid hormones, gonadotropin and ovarian carcinogenesis: A review of epidemiological and experimental data.* Gynecological Endocrinology, 2004. 19(4): p. 216-228.
13. Clinton, G.M. and W. Hua, *Estrogen action in human ovarian cancer.* Critical Reviews in Oncology/Hematology, 1997. 25(1): p. 1-9.
14. Choi, J.H., A.S.T. Wong, H.F. Huang, and P.C.K. Leung, *Gonadotropins and ovarian cancer.* Endocrine Reviews, 2007. 28(4): p. 440-461.
15. Purdie, D.M., C.J. Bain, V. Siskind, P.M. Webb, and A.C. Green, *Ovulation and risk of epithelial ovarian cancer.* International Journal of Cancer, 2003. 104(2): p. 228-32.
16. Risch, H.A., *Hormonal etiology of epithelial ovarian cancer, with a hypothesis concerning the role of androgens and progesterone.* Journal of the National Cancer Institute, 1998. 90(23): p. 1774-1786.
17. Heller, D.S., P. Murphy, and C. Westhoff, *Are germinal inclusion cysts markers of ovulation?* Gynecologic Oncology, 2005. 96(2): p. 496-9.
18. Lukanova, A. and R. Kaaks, *Endogenous hormones and ovarian cancer: epidemiology and current hypotheses.* Cancer Epidemiology Biomarkers and Prevention, 2005. 14(1): p. 98-107.
19. Key, T.J., N.E. Allen, P.K. Verkasalo, and E. Banks, *Energy balance and cancer: The role of sex hormones.* Proceedings of the Nutrition Society, 2001. 60(1): p. 81-89.
20. Rodriguez, G.C., D.K. Walmer, M. Cline, H. Krigman, B.A. Lessey, R.S. Whitaker, et al., *Effect of progestin on the ovarian epithelium of macaques: cancer prevention through apoptosis?* Journal of the Society for Gynecologic Investigation, 1998. 5(5): p. 271-6.
21. Murdoch, W. and A. McDonnel, *Roles of the ovarian surface epithelium in ovulation and carcinogenesis.* Reproduction, 2002. 123(6): p. 743-750.
22. Rodriguez, G.C., N.P. Nagarsheth, K.L. Lee, R.C. Bentley, D.K. Walmer, M. Cline, et al., *Progestin-induced apoptosis in the Macaque ovarian epithelium: differential regulation of transforming growth factor-beta.* Journal of the National Cancer Institute, 2002. 94(1): p. 50-60.
23. Henderson, B.E., R.K. Ross, M.C. Pike, and J.T. Casagrande, *Endogenous hormones as a major factor in human cancer.* Cancer Research, 1982. 42(8): p. 3232-3239.
24. Syed, V., G. Ulinski, S.C. Mok, G.K. Yiu, and S.M. Ho, *Expression of gonadotropin receptor and growth responses to key reproductive hormones in normal and malignant human ovarian surface epithelial cells.* Cancer Research, 2001. 61(18): p. 6768-6776.
25. Hildebrand, J.S., S.M. Gapstur, H.S. Feigelson, L.R. Teras, M.J. Thun, and A.V. Patel, *Postmenopausal hormone use and incident ovarian cancer: associations differ by regimen.* International Journal of Cancer, 2010. 127(12): p. 2928-2935.
26. Pearce, C.L., K. Chung, M.C. Pike, and A.H. Wu, *Increased ovarian cancer risk associated with menopausal estrogen therapy is reduced by adding a progestin.* Cancer, 2009. 115(3): p. 531-539.
27. Ho, S.M., *Estrogen, progesterone and epithelial ovarian cancer.* Reproductive Biology and Endocrinology, 2003. 1(73).
28. Schorge, J.O., J.I. Schaffer, L.M. Halvorson, B.L. Hoffman, K.D. Bradshaw, and F.G. Cunnungham, *Williams gynecology*. 2008, New York: McGraw-Hill.
29. Whiteman, D.C., M.F. Murphy, L.S. Cook, D.W. Cramer, P. Hartge, P.A. Marchbanks, et al., *Multiple births and risk of epithelial ovarian cancer.* Journal of the National Cancer Institute, 2000. 92(14): p. 1172-7.
30. Milne, R., A. Osorio, T. Ramón Y Cajal, M. Baiget, A. Lasa, E. Diaz-rubio, et al., *Parity and the risk of breast and ovarian cancer in BRCA1 and BRCA2 mutation carriers.* Breast Cancer Research and Treatment, 2010. 119(1): p. 221.
31. Cooper, G.S., J.M. Schildkraut, A.S. Whittemore, and P.A. Marchbanks, *Pregnancy recency and risk of ovarian cancer.* Cancer Causes and Control, 1999. 10(5): p. 397-402.
32. Whiteman, D.C., V. Siskind, D.M. Purdie, and A.C. Green, *Timing of pregnancy and the risk of epithelial ovarian cancer.* Cancer Epidemiology

Biomarkers and Prevention, 2003. 12(1): p. 42-46.
33. Soegaard, M., A. Jensen, E. Hogdall, L. Christensen, C. Hogdall, J. Blaakaer, et al., *Different risk factor profiles for mucinous and nonmucinous ovarian cancer: results from the Danish MALOVA study.* Cancer Epidemiology, Biomarkers and Prevention, 2007. 16(6): p. 1160-6.
34. Chen, V.W., B. Ruiz, J.L. Killeen, T.R. Coté, X.C. Wu, C.N. Correa, et al., *Pathology and classification of ovarian tumors.* Cancer, 2003. 97(S10): p. 2631-2642.
35. Kaku, T., S. Ogawa, Y. Kawano, Y. Ohishi, H. Kobayashi, T. Hirakawa, et al., *Histological classification of ovarian cancer.* Medical Electron Microscopy, 2003. 36(1): p. 9-17.
36. Folkins, A.K., E.A. Jarboe, M.H. Roh, and C.P. Crum, *Precursors to pelvic serous carcinoma and their clinical implications.* Gynecologic Oncology, 2009. 113(3): p. 391-396.
37. Kurman, R.J. and I.M. Shih, *Molecular pathogenesis and extraovarian origin of epithelial ovarian cancer-shifting the paradigm.* Human Pathology, 2011. 42(7): p. 918-931.
38. Dubeau, L., *The cell of origin of ovarian epithelial tumours.* Lancet Oncology, 2008. 9(12): p. 1191-1197.
39. Levanon, K., C. Crum, and R. Drapkin, *New insights into the pathogenesis of serous ovarian cancer and its clinical impact.* Journal of Clinical Oncology, 2008. 26(32): p. 5284-5293.
40. Seidman, J.D., P. Zhao, and A. Yemelyanova, *"Primary peritoneal" high-grade serous carcinoma is very likely metastatic from serous tubal intraepithelial carcinoma: assessing the new paradigm of ovarian and pelvic serous carcinogenesis and its implications for screening for ovarian cancer.* Gynecologic Oncology, 2011. 120(3): p. 470-473.
41. Lee, Y., F. Medeiros, D. Kindelberger, M.J. Callahan, M.G. Muto, and C.P. Crum, *Advances in the recognition of tubal intraepithelial carcininoma; applications to cancer screening and the pathogenesis of ovarian cancer.* Advances in Anatomic Pathology, 2006. 13(Number 1): p. 1-7.
42. Lee, Y., A. Miron, R. Drapkin, M. Nucci, F. Medeiros, A. Saleemuddin, et al., *A candidate precursor to serous carcinoma that originates in the distal fallopian tube.* The Journal of pathology, 2007. 211(1): p. 26-35.
43. Hesketh, R., *Introduction to cancer biology*. 2013, New York: Cambridge University Press.
44. Folkins, A.K., E.A. Jarboe, A. Saleemuddin, Y. Lee, M.J. Callahan, R. Drapkin, et al., *A candidate precursor to pelvic serous cancer (p53 signature) and its prevalence in ovaries and fallopian tubes from women with BRCA mutations.* Gynecologic Oncology, 2008. 109(2): p. 168-173.
45. Karst, A.M. and R. Drapkin, *Ovarian cancer pathogenesis: a model in evolution.* Journal of Oncology, 2010. 2010: p. 932371.
46. Baldi, A., M. Campioni, and P.G. Signorile, *Endometriosis: pathogenesis, diagnosis, therapy and association with cancer (review).* Oncology Reports, 2008. 19(4): p. 843-846.
47. Merritt, M.A., A.C. Green, C.M. Nagle, and P.M. Webb, *Talcum powder, chronic pelvic inflammation and NSAIDs in relation to risk of epithelial ovarian cancer.* International Journal of Cancer, 2008. 122(1): p. 170-176.
48. Cibula, D., M. Widschwendter, M. Zikan, and L. Dusek, *Underlying mechanisms of ovarian cancer risk reduction after tubal ligation.* Acta Obstetricia et Gynecologica Scandinavica, 2011. 90(6): p. 559-563.
49. King, S.M., T.S. Hilliard, L.Y. Wu, R.C. Jaffe, A.T. Fazleabas, and J.E. Burdette, *The impact of ovulation on fallopian tube epithelial cells: evaluating three hypotheses connecting ovulation and serous ovarian cancer.* Endocrine-Related Cancer, 2011. 18(5): p. 627-42.
50. Saleemuddin, A., A.K. Folkins, L. Garrett, J. Garber, M.G. Muto, C.P. Crum, et al., *Risk factors for a serous cancer precursor ("p53 signature") in women with inherited BRCA mutations.* Gynecologic Oncology, 2008. 111(2): p. 226-32.
51. Li, J., N. Abushahin, S.J. Pang, L. Xiang, S.K. Chambers, O. Fadare, et al., *Tubal origin of 'ovarian' low-grade serous carcinoma.* Modern Pathology, 2011. 24(11): p. 1488-1499.
52. Pearce, C.L., C. Templeman, M. Rossing, Anne, A. Lee, A.M. Near, and P. Webb, M, *Association between endometriosis and risk of histological subtypes of ovarian cancer: a pooled analysis of case-control studies.* Lancet Oncology, 2012. 13(4): p. 385-394.

53. Potter, J.D., *Morphogens, morphostats, microarchitecture and malignancy.* Nature Reviews Cancer, 2007. 7(6): p. 464-474.
54. Nezhat, F.R., R. Apostol, C. Nezhat, and T. Pejovic, *New insights in the pathophysiology of ovarian cancer and implications for screening and prevention.* American Journal of Obstetrics and Gynecology, 2015. 213(3): p. 262-267.
55. Gadducci, A., C. Sergiampietri, and R. Tana, *Alternatives to risk-reducing surgery for ovarian cancer.* Annals of Oncology, 2013. 24(suppl 8): p. viii47-viii53.
56. Merritt, M.A., M. De Pari, A.F. Vitonis, L.J. Titus, D.W. Cramer, and K.L. Terry, *Reproductive characteristics in relation to ovarian cancer risk by histologic pathways.* Human Reproduction, 2013. 28(5): p. 1406-1417.
57. Poole, E.M., M.A. Merritt, S.J. Jordan, H.P. Yang, S.E. Hankinson, Y. Park, et al., *Hormonal and reproductive risk factors for epithelial ovarian cancer by tumor aggressiveness.* Cancer Epidemiology Biomarkers and Prevention, 2013. 22(3): p. 429-436.