Ovary and Peritoneal Washings

11

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Contents

Abbreviations

AFP	Alpha-fetoprotein
AGCT	Adult granulosa cell tumor
AJCC	American Joint Committee on Cancer
BSO	Bilateral salpingo-oophorectomy
CEA	Carcinoembryonic antigen
E2	Estradiol
ER	Estrogen receptor
FIGO	International Federation of Gynecology and Oncologists
FNA	Fine-needle aspiration
hCG	Human chorionic gonadotropin
HLM	Hemosiderin-laden macrophage
IHC	Immunohistochemistry
JGCT	Juvenile granulosa cell tumor
PR	Progesterone receptor
WHO	World Health Organization

List of Frequently Asked Questions:

Ovary

1. What are the indications for fine-needle aspiration of the ovary?

Fine-needle aspiration (FNA) of the ovary was first described in the early 1970s [1, 2]. The relative number of indications for ovarian FNA is limited but growing. Aspiration of the ovary is technically similar to other abdominal sites, and it is relatively safe and inexpensive. FNA of an ovarian mass is used in the following clinical scenarios:

- Diagnosis and therapy of a persistent ovarian mass in women of reproductive age [3].
- Drainage of extremely large benign-appearing cysts to allow for laparoscopic removal [4].
- Avoidance surgical intervention during pregnancy [5].
- Evaluation of malignancy in patients with a prior diagnosis or treated cancer, particularly in cases where laparoscopy is contraindicated.

For the cytologist, it is important to appreciate that a surgical approach is generally recommended for complex or solid lesions of the ovary. However, many patients present with a pelvic mass of unknown origin, and these occult ovarian lesions are often sampled by FNA. Ovarian FNA is contraindicated in the setting of acute abdominal/pelvic pain, as the procedure may delay treatment of serious conditions, such as torsion [6].

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2. How are ovarian FNA samples obtained?
Aspiration can be performed transvaginally, transrectally, laparoscopically, at the time of laparotomy, or percutaneously through the abdomen, with or without imaging guidance. It is important to note the route of aspiration because contamination with normal tissue can occur and be somewhat problematic.

3. Why is aspiration of the ovary uncommon?
In the modern setting, ovarian aspiration is limited to a few circumstances, largely due to three main considerations:

1. There is concern that FNA of a malignant cyst can lead to tumor spillage and induce peritoneal seeding [7].
2. Aspiration as a therapeutic technique is not useful because a high percentage (up to 75%) of benign cysts will recur and ultimately require excision [8–10].
3. The diagnostic accuracy of ovarian FNA is controversial, and many malignant lesions are missed, especially in premenopausal patients [11].

As an additional concern, ovarian aspiration results in a high rate of unsatisfactory diagnoses (up to 20%), and ultrasound is as good or better at determining the malignant potential of ovarian lesions [12]. For cases in which radiologic imaging suggests malignancy, clinicians will recommend up-front surgery to evaluate an ovarian lesion, circumventing the need for FNA.

4. How accurate is ovarian FNA in the diagnosis of malignancy?
The answer to this question is somewhat unclear because various studies have defined the sensitivity and specificity in different ways. Although some report high values for sensitivity and specificity (up to 84% and 93%, respectively) [1, 13], these values are likely exaggerated because borderline tumors were excluded from analysis. Borderline tumors are a common cause of false-negative FNAs, because the cyst fluid is relatively acellular, perhaps due to greater cell-to-cell adhesions than their malignant counterparts. If borderline tumors are included in the analysis, the sensitivity ranges from 26% to 54% [14, 15]. Even though it is generally agreed that the test has a high specificity (>90%), the low sensitivity is a valid criticism of the technique because it limits the primary usefulness of the technique – as a rule out test for carcinoma.

5. What is the risk of iatrogenic peritoneal seeding following ovarian FNA?
Aspiration of ovarian cysts is considered taboo by some clinicians due to the risk of seeding an early stage ovarian cancer. However, the actual rate of seeding is not known [11]. The issue of seeding was first raised in a single but influential article that reported two cases in which surgical resection was delayed after fine-needle aspiration [7]. Iatrogenic peritoneal seeding was not confirmed in either case, but a strong argument was made that FNA has the potential to delay treatment, which may result in a worse prognosis for patients who already had peritoneal disease or cyst rupture prior to sampling. While seeding risk may in fact be minimal, the effect of malignant cyst rupture before and during surgery in patients with early stage ovarian cancer has been evaluated. One study of 60 patients with stage I epithelial ovarian carcinoma showed cyst rupture *during* surgery had no influence on survival rates (average follow-up 75 months) [16], with similar results shown in other studies as well [17, 18]. However, in patients who had cysts ruptured *prior to* surgery, there was a significant survival difference (10-year survival of 59% vs. 78% that had an intact capsule) [19]. The risk of seeding from an acute surgical spill seems to be small or nonexistent because surgeons irrigate spillage immediately. In contrast, slow and continuous spillage into the peritoneal cavity following disruption (biologic or iatrogenic) may create a favorable environment for peritoneal implantation. For this reason, ultrasound-guided FNA is considered to have more risk of a significant treatment delay than laparoscopic evaluation, which can often provide histologic information and therapy without much delay [7].

6. What are the other complications associated with ovarian FNA?
The risk of other major complications appears to be low for this procedure. In one of the largest studies, which included 893 patients, the most common complications of ovarian FNA were mild vagal symptoms (17, 2%) and transient mild-to-moderate pain (8, 0.9%) [20]. The most serious complications noted were acute abdominal pain (6, 0.7%) and infection (4, 0.4%). However, 6 (60%) of these patients required surgery, and no other life-threatening complications were reported. Severe pelvic infection following transvaginal and transrectal approaches has been seen in up to 1.3% of patients [13]. The findings of a low complication rate are similar to the complications of abdominal FNA in general [21].

7. Is there any therapeutic value to the aspiration of ovarian cysts?
Aspiration of small ovarian cysts has no apparent therapeutic benefit over other therapies. The vast majority (up to 71%) of ovarian cysts regress after a short-term course of oral contraceptives [22]. Of those masses that do not regress (endometriomas, benign neoplasia, benign para-ovarian cysts, hydrosalpinx, and malignant tumors), only para-ovarian cysts are appropriately treated by aspiration. As a rule of thumb, the larger the ovarian cyst, the greater the risk of recurrence [12]. Of note, endometriomas are generally not

acceptable to aspirate, as the underlying endometriosis ensures that the cysts will recur [3].

8. What are the ultrasonographic features of benign versus malignant ovarian lesions?

Benign and malignant ovarian masses demonstrate characteristic features on ultrasound. The following is a list from a comprehensive study of 211 adnexal masses (183 benign and 28 malignant) [23]. The authors found the following features associated with benign ovarian lesions:

- No solid component (54% benign vs. 0% malignant, $p < 0.001$)
- If present, a hyperechoic solid component (15% benign vs. 0% malignant, $p < 0.001$)
- An echogenic fluid component (58% benign vs. 21% malignant, $p < 0.001$)
- Thin (<3 mm) septations (26% benign vs. 4% malignant, $p = 0.02$)
- Thin (<3 mm) wall (50% benign vs. 29% malignant, $p < 0.001$)
- Normal free fluid in the abdomen (98% benign vs. 68% malignant, $p < 0.001$)
- Peripheral only or no flow detected by Doppler (83% benign vs. 14% malignant, $p < 0.001$)

In contrast, malignant ovarian lesions demonstrated the following ultrasonographic features:

- Nonhyperechoic solid component (32% benign vs. 100% malignant $p < 0.001$)
- An echogenic or no fluid component (43% benign vs. 61% malignant, $p < 0.001$)
- Thick (≥3 mm) septations (17% benign vs. 14% malignant, $p = 0.02$)
- Thick (≥3 mm) wall (43% benign vs. 32% malignant, $p < 0.001$)
- Abnormal amount of free fluid in the abdomen (2% benign vs. 32% malignant, $p < 0.001$)
- Central flow by Doppler (17% benign vs. 86% malignant, $p < 0.001$)

Using the above criteria, the authors developed a scoring formula with a sensitivity of 93% and specificity of 93% [23], which demonstrates why radiographic imaging is routinely used to identify lesions that require surgical evaluation.

Of note, some ultrasound findings are surprisingly *not* useful to predict a benign or malignant diagnosis ($p > 0.05$), including:

- Unilaterality (86% benign vs. 71% malignant)
- Bilaterality (7% benign vs. 14% malignant)
- Average size (5.1 cm benign vs. 5.9 cm malignant)
- Maximum size (6.0 cm benign vs. 6.9 cm malignant)

9. What is the role of ancillary testing to diagnose ovarian lesions?

Specimens obtained from ovarian aspiration are generally cyst fluids, which should be sent to the clinical laboratory for marker assessment. Measurements of estradiol (E2), CA-125, carcinoembryonic antigen (CEA), and alpha-fetoprotein (AFP) can be useful. E2 is a compound that is elevated in functional cysts but absent in epithelial lesions [24, 25]. CA-125 and CEA can be useful as tumor markers but are generally considered to be nonspecific. An elevated AFP is commonly associated with germ cell tumors of the ovary but can also elevated in teratomas, sex cord-stromal tumors, and other ovarian epithelial neoplasms.

10. What are the adequacy criteria for an ovarian FNA?

A nondiagnostic interpretation can be rendered in cases of low cellularity or in cases of poorly preserved cells. Although strict adequacy has not been established for ovarian FNA, studies have used the criteria of at least six groups of epithelial cells to make a diagnosis [11].

11. What cellular components are found in aspirates from normal ovaries?

- **Ovarian Stroma**: Fragments of normal ovarian stroma will appear as cohesive groups of small spindle cells containing elongated nuclei with blunt or tapered ends. Ovarian fibromas will have a similar cytologic appearance to normal ovarian stroma, although they are unlikely to yield much cellular material due to dense (and often abundant) intercellular collagen.
- **Germinal Epithelium:** If follicles are aspirated, germinal epithelium will appear as flat sheets of epithelioid cells with oval nuclei, indistinct nucleoli, and a small-to-moderate amount of watery cytoplasm.
- **Simple Nonfunctional Cysts:** Unilocular cysts are a normal finding and often diagnosed at resection as cortical inclusion cysts, paratubal serous cysts, hydrosalpinx, or benign simple cysts. These cysts are indistinguishable by cytology, yielding small groups of cuboidal epithelial cells accompanied by foamy histiocytes. These cysts are considering "nonfunctional," meaning that normal hormonal cycles do not influence their growth.
- **Cystic Follicles and Follicular Cysts:** Cystic follicles and follicular cysts differ only in the size of the lesion, with follicles larger than ~2 cm designated as follicular cysts. Follicular lesions may represent a potential pitfall for cytologists, especially since ovarian FNAs are an uncommon specimen. Follicular cysts are benign physiologic (or "functional") cysts composed of two cell types; an inner

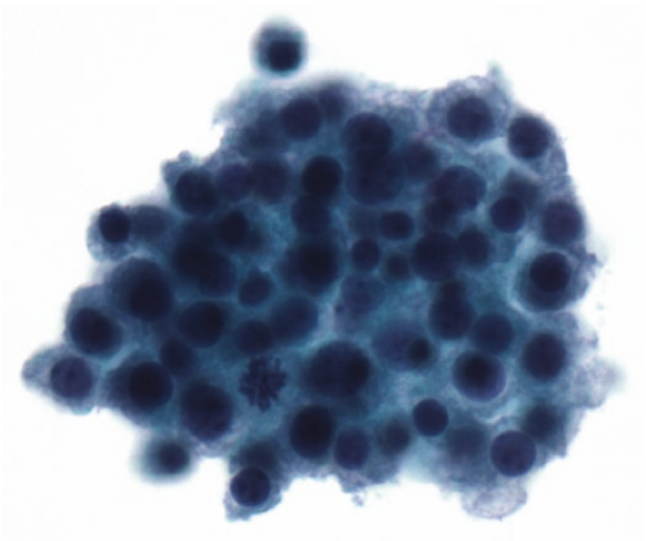

Fig. 11.1 Follicular cyst. Granulosa cells have granular and occasionally vacuolated cytoplasm and a large round-to-oval nucleus. These cells can appear immature, pyknotic, and mitotically active, mimicking malignancy (ThinPrep, Papanicolaou stain)

layer of granulosa cells overlying an outer layer of theca cells. Fluid from follicular cysts can be hypercellular, with the cells having granular and occasionally vacuolated cytoplasm and a large round-to-oval nucleus. The cells can appear immature, and mitotic figures are invariably present (Fig. 11.1); thus, it is not entirely surprising that follicular cysts are a common cause of false-positive results [26, 27]. The differential diagnosis includes granulosa cell tumor, carcinoid tumor, and serous neoplasia. Follicular cysts are composed of granulosa cells, so granulosa cell tumor should be excluded. Follicular cysts contain luteinized or nonluteinized granulosa cells, with the luteinized cells containing more abundant cytoplasm and larger nuclei.

- **Corpora Lutea and Corpus Luteum Cysts:** These functional cysts yield cellular aspirates with luteinized granulosa cells, appearing singly and in small clusters. These cells are epithelioid with round-to-oval eccentric nuclei and fine chromatin with prominent nucleoli. Luteal cells contain abundant granular cytoplasm with small vacuoles. The background may contain histiocytes, red blood cells, or hemosiderin-laden macrophages, especially in more advanced cysts. Corpus luteum cysts may arise during pregnancy and present as large lesions concerning for malignancy; aspirates may show marked cytoplasmic vacuoles or large cells with hyaline droplets [28].

12. What contaminants appear in ovarian aspirates?

Aspirates from ovarian lesions may contain contaminating epithelium from surround organs or those through which the needle passes. These include squamous epithelium indirectly sampled from transvaginal procedures and mesothelium from percutaneous abdominal aspirates. Columnar intestinal epithelium and mucus may be present if the needle pierces the intestines, and urothelial epithelium may be present if the bladder is punctured.

13. What are the diagnostic components of endometriomas (endometriotic cysts)?

Endometriomas are often referred to as "chocolate cysts" due to the characteristic thick dark brown fluid found within these lesions. The major component of endometriomas is hemosiderin-laden macrophages (HLMs) in a background of degenerated blood. Similar to endometriosis (discussed elsewhere), endometrioid cells can be found singly, in small clusters, or monolayered sheets. These cells represent ectopic endometrium, with benign round-to-oval nuclei with uniform chromatin and variable cytoplasm. Endometrial stromal cells, if present, appear as 3-dimensional cohesive clusters of spindles cells with oval nuclei and scant cytoplasm. Although most hemorrhagic cysts are endometriomas at histologic evaluation, benign and malignant neoplasia can also present in this way [3]. Thus, it is important to distinguish benign endometriomas from neoplastic hemorrhagic cysts, which also contain abundant HLMs.

14. How are benign follicular cysts distinguished from granulosa cell tumors by cytology?

Granulosa cell tumors exhibit nuclear atypia not seen in functional cysts, including pale and finely dispersed chromatin and nuclear membrane irregularities including grooves. However, nuclear grooves are not specific for granulosa cell tumor and are sometimes present in granulosa cells of functional cysts. Call-Exner bodies are frequently present in granulosa cell tumors. These are homogeneous aggregates of basement membrane surrounded by granulosa cells. Mitotic figures are not specific and can be found in either entity. Radiologic and clinical impressions of a benign cyst can be extremely helpful and reassuring.

15. What are the most common ovarian tumors?

Most ovarian tumors are benign (Table 11.1), including benign teratomas, cystadenomas, and stromal tumors, which comprise 71% of ovarian neoplasms [29].

Table 11.1 Most common ovarian tumors

Primary ovarian tumors	Frequency (%)
Benign cystic teratoma	32
Benign serous tumors	16
Benign mucinous tumors	14
Serous adenocarcinomas	9
Sex cord-stromal tumors	9
Borderline serous tumor	4
Endometrioid adenocarcinomas	3
Borderline mucinous tumor	1
Clear cell carcinoma	1
Mucinous adenocarcinoma	1

16. What are the most common histologic subtypes of epithelial ovarian tumors?

Tumor with serous histology is more common than other subtypes of epithelial ovarian tumors, followed by primary ovarian mucinous tumors (Table 11.2). Endometrioid and clear cell tumors of the ovary are relatively rare by comparison [30].

17. What are the key gross and cytologic features of benign epithelial-stromal tumors?

The most common benign epithelial-stromal tumors are serous cystadenomas. The adjective "serous" describes gynecologic epithelial tumors that appear similar to the ovarian surface and fallopian tube epithelium. Serous cystadenomas can be unilocular or multilocular, and the vast majority will contain clear fluid. The cyst wall will be smooth and may contain rounded nodules but will lack papillary excrescences. Many benign serous tumors are associated with mesenchymal stromal proliferation, and the term "-fibroma" is appended to the diagnosis to denote these lesions with a solid fibrous component (e.g., "serous cystadenofibroma").

Table 11.2 Histologic subtypes of ovarian epithelial tumors

Histologic subtype	Total (%)
Serous	46
Mucinous	36
Endometrioid	8
Clear cell	3
Other[a]	7

[a]Includes Brenner/transitional, undifferentiated, and mixed

Benign serous cysts contain cuboidal cyst-lining cells. In the absence of atypical features, ciliated cells with terminal bars, apical cytoplasm, and basal nuclei are diagnostic of benign serous cysts (Fig. 11.2). Psammomatous calcifications can be present. Benign-appearing spindle cells will be evident if the needle has sampled a solid fibrous component or normal ovarian stroma.

Brenner tumors are epithelial-stromal neoplasms that contain transitional (urothelial) type epithelium in a fibrous stroma. Benign Brenner tumors are typically solid and unilateral with a smooth cut surface. They can have microcysts, large cysts, or be associated with other benign (or malignant) ovarian neoplasms. Rarely, Brenner tumors can have borderline or malignant features (<1% of cases). The cytology of Brenner tumors can be difficult because the epithelial component can yield hypercellular aspirates whereas the fibromatous component may not sample well (Fig. 11.3). However, the epithelial cells are generally bland and polygonal with a generous amount of cytoplasm, and mitotic figures are rare. Brenner tumor epithelium is arranged in whorled nests inside fibrous stroma, a characteristic feature that make a cell block or concurrent core biopsy particularly helpful for the diagnosis.

18. How often are serous neoplasms benign?

Approximately, 50% of all serous tumors are benign at resection, and diagnostic entities include serous cystadenoma, serous adenofibroma, and serous cystadenofibroma (Table 11.3) [31]. Borderline tumors only comprise a small proportion of serous neoplasia.

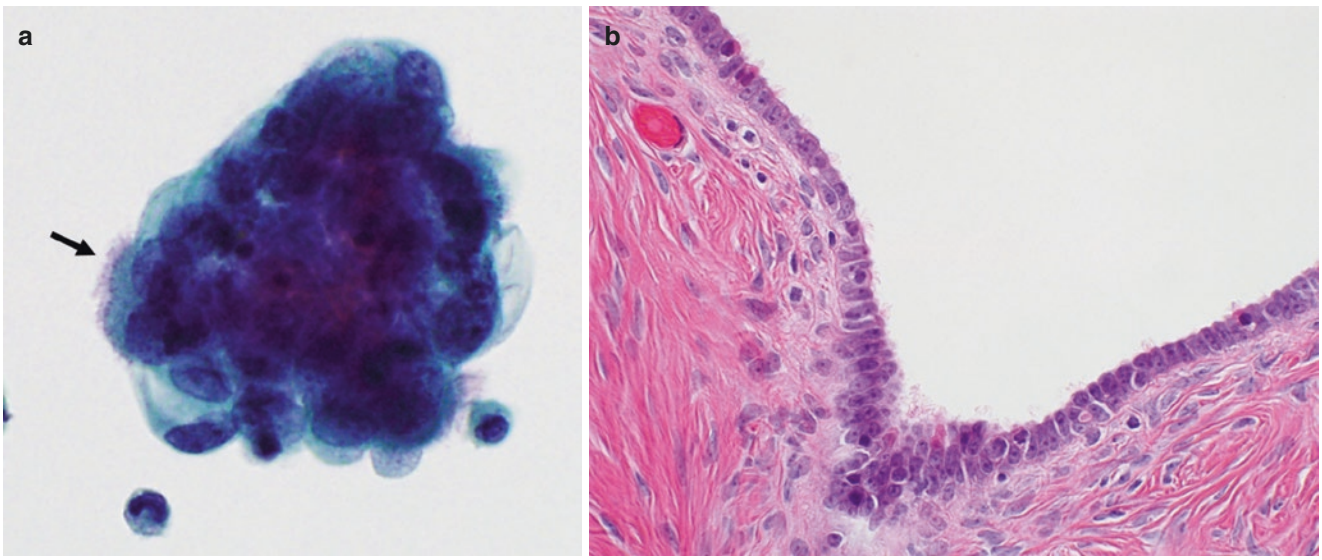

Fig. 11.2 Ovarian serous cystadenofibroma. (**a**) Groups of benign-appearing cells with cilia are typically seen in serous cystadenoma (ThinPrep, Papanicolaou stain). (**b**) Tumor demonstrates a simple lining of cuboidal epithelium overlying a dense fibrous component on resection. Note that many of the cyst-lining cells are ciliated (H&E stain)

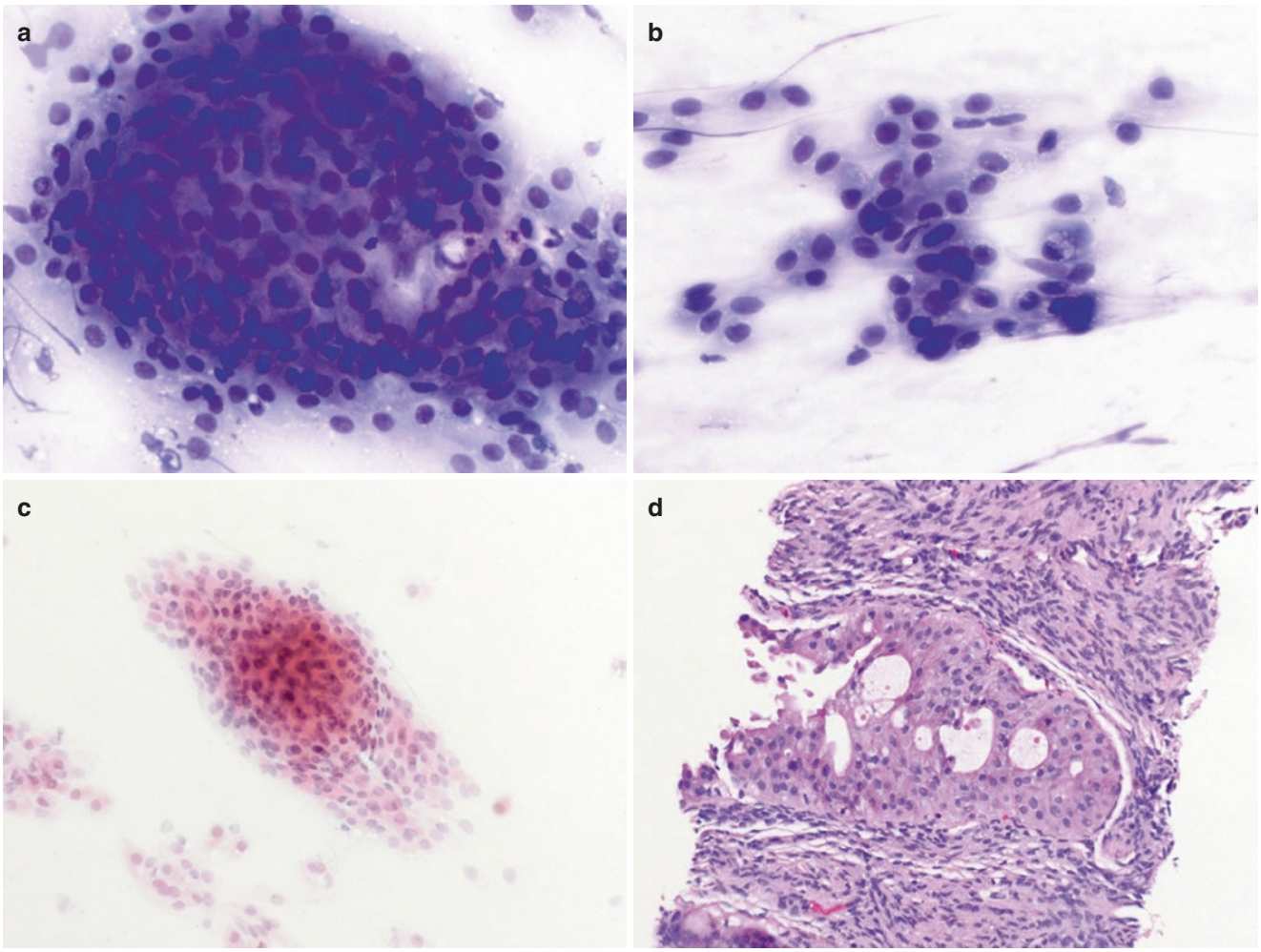

Fig. 11.3 Brenner tumor. (**a**) FNA of an ovarian Brenner tumor (air-dried smear) demonstrates whorled groups of benign-appearing cells with abundant cytoplasm (Diff-Quik stain). (**b**) Brenner tumors can be hypercellular, but the cells lack malignant features like mitotic figures and necrosis (Diff-Quik stain). (**c**) Alcohol-fixed preparation showing a whorled group of Brenner tumor cells (Papanicolaou stain). (**d**) Core biopsy demonstrates a rounded whorled group of epithelial cells in a background of cellular stroma (H&E stain)

Table 11.3 Histologic diagnoses of serous epithelial ovarian tumors

Diagnosis	Total (%)
Benign serous cysts	50
Serous borderline tumor	15
Serous adenocarcinoma	35

19. Is it possible to distinguish benign serous neoplasms, borderline serous tumors, and low-grade serous adenocarcinomas by cytology?

It is not possible to distinguish serous borderline tumors from low-grade serous adenocarcinomas by gross or cytologic examination. The histologic diagnosis of serous adenocarcinoma requires the presence of stromal invasion. For this reason, the term "low-grade serous neoplasia" is often used to describe lesions that have the cytologic appearance of borderline and low-grade serous adenocarcinoma. However, aspirates of serous borderline tumors (Fig. 11.4) will generally have less atypia and cellularity than those of low-grade serous adenocarcinomas (Fig. 11.5). The presence of nuclear atypia distinguishes low-grade serous tumors from benign serous cystadenomas, which exhibit bland and ciliated epithelium.

20. What are the key cytologic and immunohistochemical features of benign and malignant serous neoplasms?

It is not always possible to distinguish benign from malignant serous neoplasms by cytology, but there are some features common to either case (Table 11.4). Borderline neoplasms are tumors with uncertain malignant potential that still have a favorable prognosis even if they recur. Psammomatous calcifications are common in both serous borderline tumors and low-grade serous adenocarcinomas, found in approximately one-third of cases. Serous carcinomas are graded using a two-tier system that highly correlates

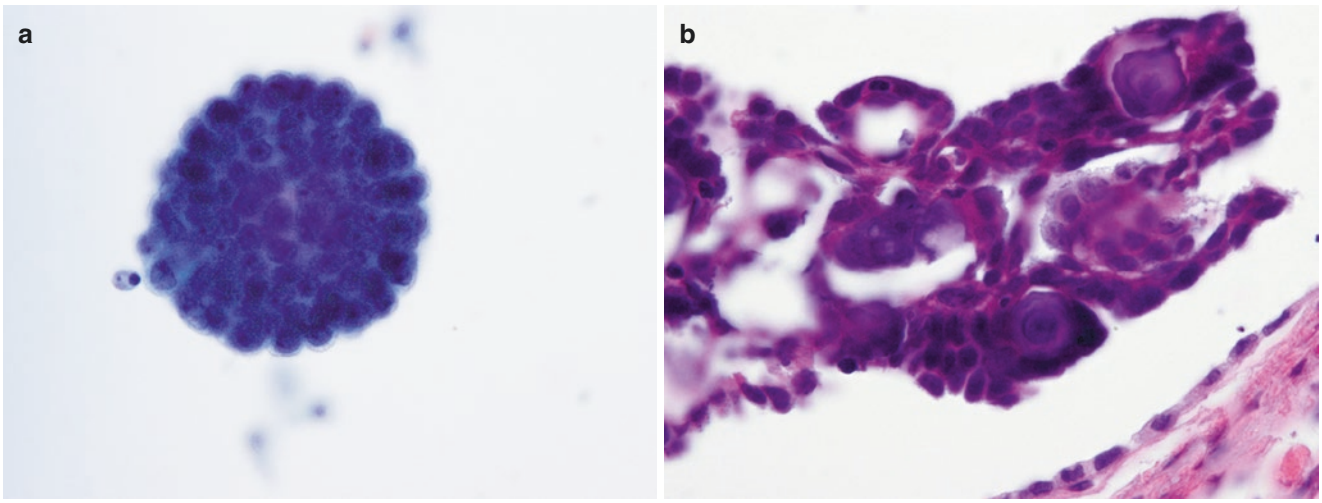

Fig. 11.4 Serous borderline tumor. (**a**) FNA of serous borderline tumors may be hypocellular with few cell groups, but lesional tissue should not demonstrate significant cytologic or nuclear atypia (Papanicolaou stain). In contrast to serous cystadenomas, cells do not have cilia. (**b**) Cell block preparation of the FNA demonstrated small strips of epithelial cells associated with psammoma bodies (H&E stain)

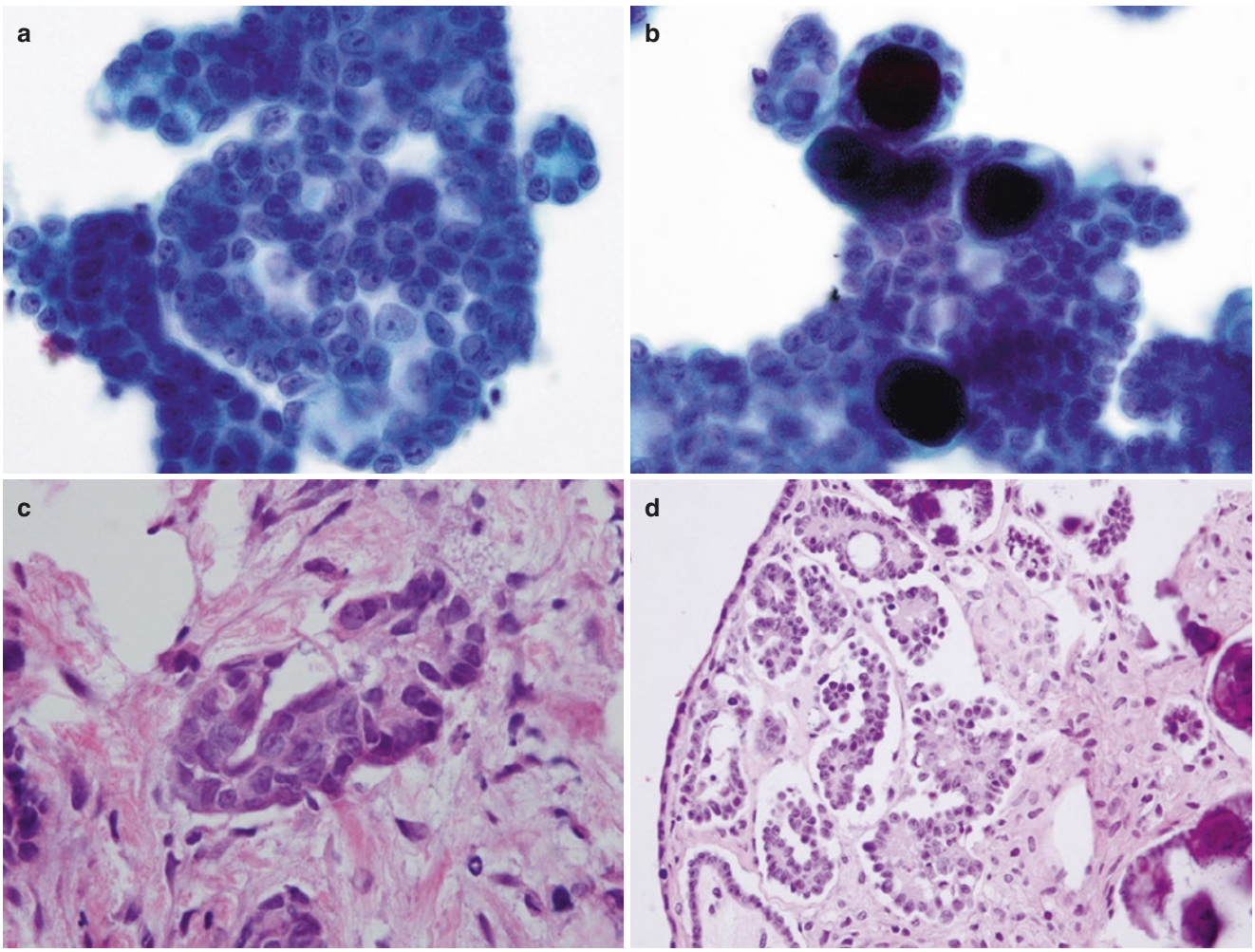

Fig. 11.5 Low-grade serous adenocarcinoma. (**a**) In contrast to serous cystadenomas and borderline tumors, this aspirate of a low-grade serous adenocarcinoma shows a crowded sheet of cells with enlarged irregular nuclei. (**b**) Psammoma bodies are frequent in low-grade serous adenocarcinomas. (**c**) Cell block preparations may reveal foci concerning for invasion. (**d**) Retraction artifact is commonly observed in low-grade serous adenocarcinomas and may be apparent in cell block fragments (**a**, **b**: Papanicolaou stain; **c**, **d**: H&E stain)

with long-term prognosis [32]. Low-grade serous carcinoma will appear in small clusters or in crowded sheets. Cytoplasmic vacuoles may be present, and cells will exhibit an increased nuclear: cytoplasmic ratio, with somewhat irregular nuclei and prominent nucleoli. High-grade tumors are more likely to yield positive washings than low-grade tumors and have frankly malignant cytology (Fig. 11.6) [33].

Table 11.4 Cytologic features of serous neoplasms

Benign serous tumors	Malignant serous tumors
Sparsely cellular	Higher cellularity associated with higher grade tumors; serous borderline tumors may be sparsely cellular and falsely negative
Bland, columnar, ciliated epithelium	Enlarged crowded cells with overlapping nuclei; irregular nuclear membranes; prominent nucleoli
Relatively clean background, some histiocytes; psammoma bodies rare	Psammoma bodies present in 30%
DDx includes cystadenoma, cystadenofibroma, cortical inclusion cyst, paratubal cysts, and hydrosalpinx	DDx includes serous borderline tumor and low-grade serous adenocarcinoma; overtly malignant cytologic features may indicate high-grade serous carcinoma

Serous neoplasms have characteristic immunohistochemical features that are sometimes essential for the diagnosis, especially when considering other tumor subtypes and metastatic lesions. Serous lesions are typically CK7 positive and CK20 negative, which can be helpful for distinguishing these from colorectal malignancies. Additionally, they are negative for CDX-2 and express PAX-8. WT-1 is a helpful positive marker because it distinguishes serous carcinoma from endometrioid adenocarcinomas and clear cell carcinomas of the ovary. Estrogen receptor (ER) and progesterone receptor (PR) exhibit variable positivity in serous carcinomas but are more likely expressed in low-grade neoplasms. In contrast to low-grade serous neoplasia, high-grade serous carcinomas almost always have a mutant (overexpression or loss) p53 phenotype and are diffusely and strongly reactive for p16.

21. What is the significance of p53 immunohistochemistry in high-grade serous carcinoma? How do I report p53 staining results?

The tumor suppressor gene, *TP53*, is mutated in ~96% of tubo-ovarian high-grade serous carcinomas [34], and immunohistochemistry for the protein product, p53, can act as a surrogate marker of *TP53* mutation status (Fig. 11.7).

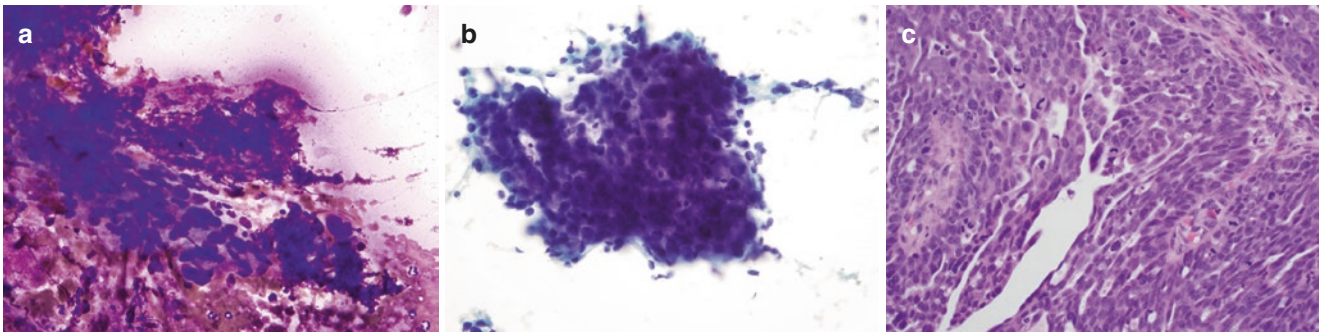

Fig. 11.6 High-grade serous carcinoma of the ovary. FNA smears demonstrate high-grade epithelial cells associated with necrosis (**a**: Diff-Quik stain; **b**: Papanicolaou stain). (**c**) Characteristic features of high-grade serous tumors include solid or papillary architecture, crowded overlapping cells, pleomorphic hyperchromatic nuclei, and abundant mitotic figures (cell block, H&E stain)

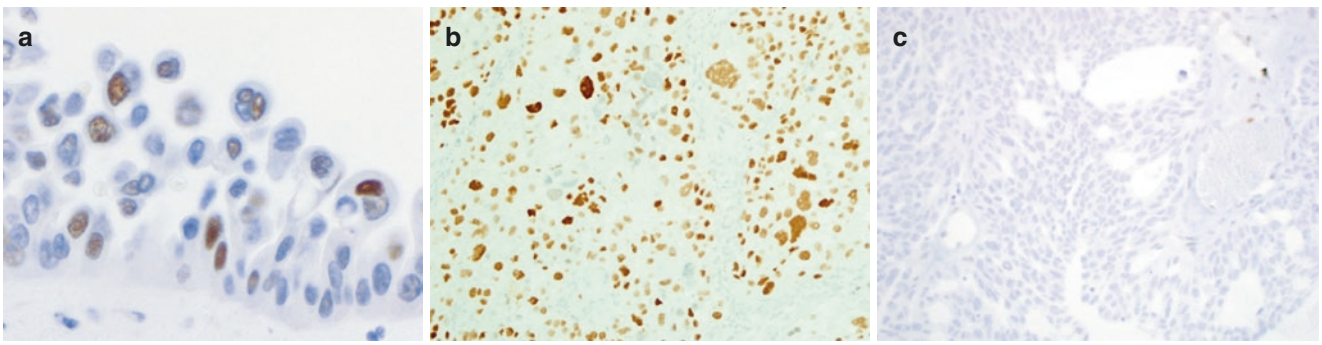

Fig. 11.7 Immunophenotypes of p53. (**a**) Wild-type staining pattern of p53 demonstrates heterogeneity, with variable strong and weak intensity seen in positive cells. (**b**) The most common mutant phenotype of p53 is a strong and diffuse staining pattern. (**c**) In a minority of cases, p53 demonstrates a null phenotype, indicating loss of the immunogenic portions of the p53 protein

Inactivating (missense) mutations of the gene result in increased nuclear expression of the p53 protein, yielding a diffusely and strongly positive staining pattern. Deleterious (nonsense and frameshift) mutations of *TP53* result in expression of a truncated protein, resulting in a null expression phenotype. The mechanism of p53 mutation is conceptually important because nonsense and frameshift mutations will still express N-terminal portions of the p53 protein, which may or may not be detectable by IHC. In one study correlating *TP53* mutation status with p53 expression, 62% had missense mutations (of which 100% had diffuse and strong p53 by IHC), 16% had nonsense mutations (of which 55% exhibited a null expression phenotype), and 13% had frameshift mutations (78% of which showed a null phenotype by IHC) [35].

If no mutations of *TP53* are present, p53 will display a wild-type staining pattern with weak expression in a heterogeneous distribution. Therefore, we recommend that p53 immunohistochemistry generally be reported in one of the following ways:

- POSITIVE p53 (diffuse and strong expression, mutant phenotype).
- POSITIVE p53 (heterogeneous expression, wild-type phenotype).
- NEGATIVE p53 (null expression, mutant phenotype).

22. How often is p53 mutated in gynecologic malignancies?

When confronted with adnexal masses and/or peritoneal implants, there is often a question of whether the tumor originated from the ovary, the uterus, or the peritoneum (the latter presumably arises from endometriosis or benign Müllerian inclusions). For the most part, the answer to this question is addressed after resection by surgical pathology. This is particularly true for primary peritoneal Müllerian disease because the diagnosis requires exclusion of ovarian and uterine primaries. However, p53 is a useful tool for separating low-grade from high-grade neoplasms, as well as high-grade serous from ovarian tumors with endometrioid and clear cell histology. This utility is best illustrated by examining the rate of p53 mutations in each of the tumor subtypes. Somewhat surprisingly, the p53 mutations occur at slightly different frequencies depending on the origin.

Primary Ovarian Tumors:

- 8% of borderline serous tumors have p53 mutations [36].
- 8% of low-grade serous carcinomas have p53 mutations [36].
- 96% of high-grade serous carcinomas have p53 mutations [34].
- 22% of endometrioid adenocarcinomas of the ovary have a mutant p53 immunophenotype [37].
- 0–3% of clear cell adenocarcinomas of the ovary have p53 mutations by sequencing [37, 38].

Primary Uterine Tumors:

- 90% of uterine serous carcinomas are found to have *TP53* mutations [39].
- 12% of endometrial endometrioid adenocarcinomas have a mutant p53 immunophenotype, including 40% of grade 3 tumors and only 3% of grade 1 and 2 tumors [40].
- 34% of clear cell adenocarcinomas of the uterus have a mutant p53 immunophenotype [41].
- 91% of uterine carcinosarcomas have p53 mutations (cBioPortal, TCGA provisional data).

Although useful for separating low-grade and high- grade serous tumors, p53 cannot distinguish high-grade tubo-ovarian from uterine serous carcinomas and carcinosarcomas. Only a small proportion of any endometrioid tumors will harbor p53 mutations, the vast majority of those being high grade. Clear cell carcinomas of the uterus exhibit p53 mutations more often than ovarian primaries.

23. What are the key cytologic and immunohistochemical features of endometrioid neoplasms of the ovary?

Endometrioid adenocarcinomas are usually cystic and solid tumors that are morphologically similar to endometrial endometrioid adenocarcinomas. In fact, these tumors are thought to arise from endometriosis and endometriomas, which are identified histologically in nearly half of ovarian endometrioid adenocarcinomas [31]. As seen in the endometrium, squamous differentiation is a common finding. Unlike serous and mucinous carcinomas, endometrioid borderline tumors are rarely encountered.

Cytologically, ovarian endometrioid adenocarcinomas are identical to those of the endometrium, with pseudostratified glandular cells in clusters with enlarged oval nuclei (Fig. 11.8). A cell block can be helpful to visualize well-differentiated endometrioid adenocarcinomas, but high-grade endometrioid lesions can be difficult to distinguish from other high-grade malignancies. Immunohistochemistry may be helpful, as these will typically have an immunoprofile strong positive for PAX-8, ER, and PR, patchy p16, wild-type p53 (unless high grade), and negative for WT-1.

24. What are the key cytologic and immunohistochemical features of clear cell neoplasms of the ovary?

Clear cell tumors of the ovary typically exhibit large, pleomorphic nuclei and abundant clear vacuolated cyto-

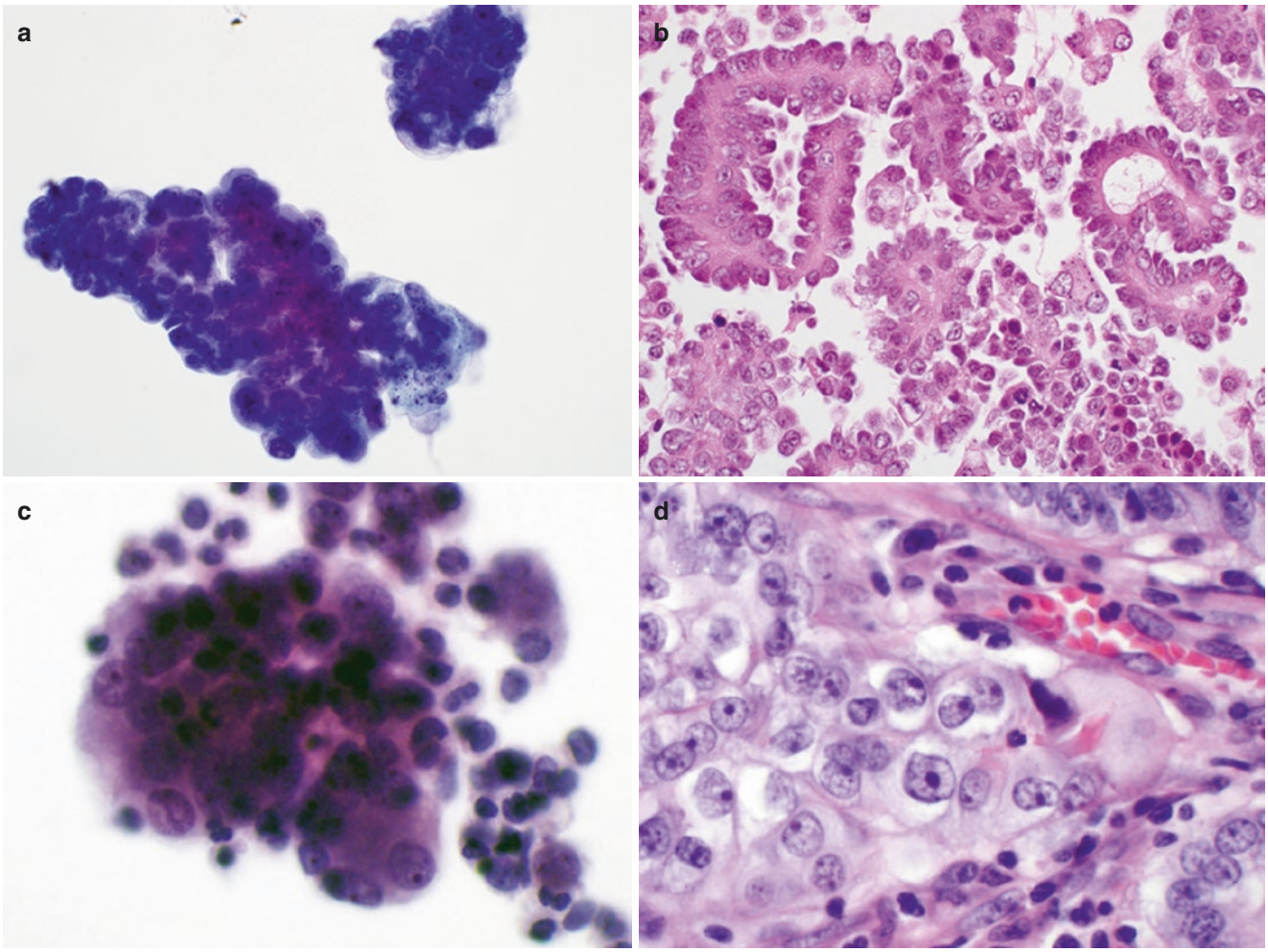

Fig. 11.8 Endometrioid adenocarcinoma of the ovary. (**a**) Peritoneal washing of a well-differentiated endometrioid adenocarcinoma of the ovary with clusters of atypical glandular cells (Papanicolaou stain). (**b**) The glandular architecture is best appreciated on the cell block sections (H&E stain). (**c**) FNA of a pelvic mass demonstrating high-grade endometrioid adenocarcinoma of the ovary (Papanicolaou stain). (**d**) Cell block preparation demonstrating high-grade pleomorphic nuclei with vesicular chromatin and prominent nucleoli (H&E stain)

plasm (Fig. 11.9). These tumors are considered high-grade malignancies, with <1% considered benign clear cell adenomas or borderline malignancies [31]. Immunohistochemistry is helpful if clear cell carcinomas are suspected, as they are typically positive for Napsin-A, AMACR, CK7, EMA, HNF1-B, wild-type for p53, and negative for ER, PR, and WT-1 [42]. Of note, clear cell carcinomas of both the ovary and kidney are positive for PAX-8, so other markers may be necessary if the primary site is uncertain.

25. What are the key cytologic and immunohistochemical features of ovarian mucinous tumors? Can they be distinguished from gastrointestinal metastasis?

Aspirates from mucinous tumors of the ovary often have a variable cytologic appearance (Fig. 11.10). Well-differentiated components may appear columnar with mucin vacuoles and exhibit only mild nuclear atypia. For this reason, it is difficult to distinguish mucinous borderline tumors from mucinous adenocarcinoma, and this is of minor concern to cytologists because as resection is required to exclude intramucosal carcinoma or invasion [43]. Mucinous ovarian tumors may cause pseudomyoxma peritonei, but the majority of adnexal mucinous tumors presenting with pseudomyxoma are in fact metastases from gastrointestinal sites. The finding of abundant mucin (in either peritoneal washings or an ovarian aspiration) is at least atypical, if not suspicious for a neoplastic process.

Mucinous adenocarcinomas of the ovary may arise from mucinous borderline tumors, and thus low-grade elements are suggestive of an ovarian primary rather than a metastasis [44]. Immunohistochemistry can be useful, as metastatic lesions from the colon and appendix will be positive for CK20, CDX-2, and SATB2 and largely negative for CK7, PAX-8, and WT-1. Often, however, a diagnosis of "mucinous cystic neoplasm" is sufficient to guide management [45, 46].

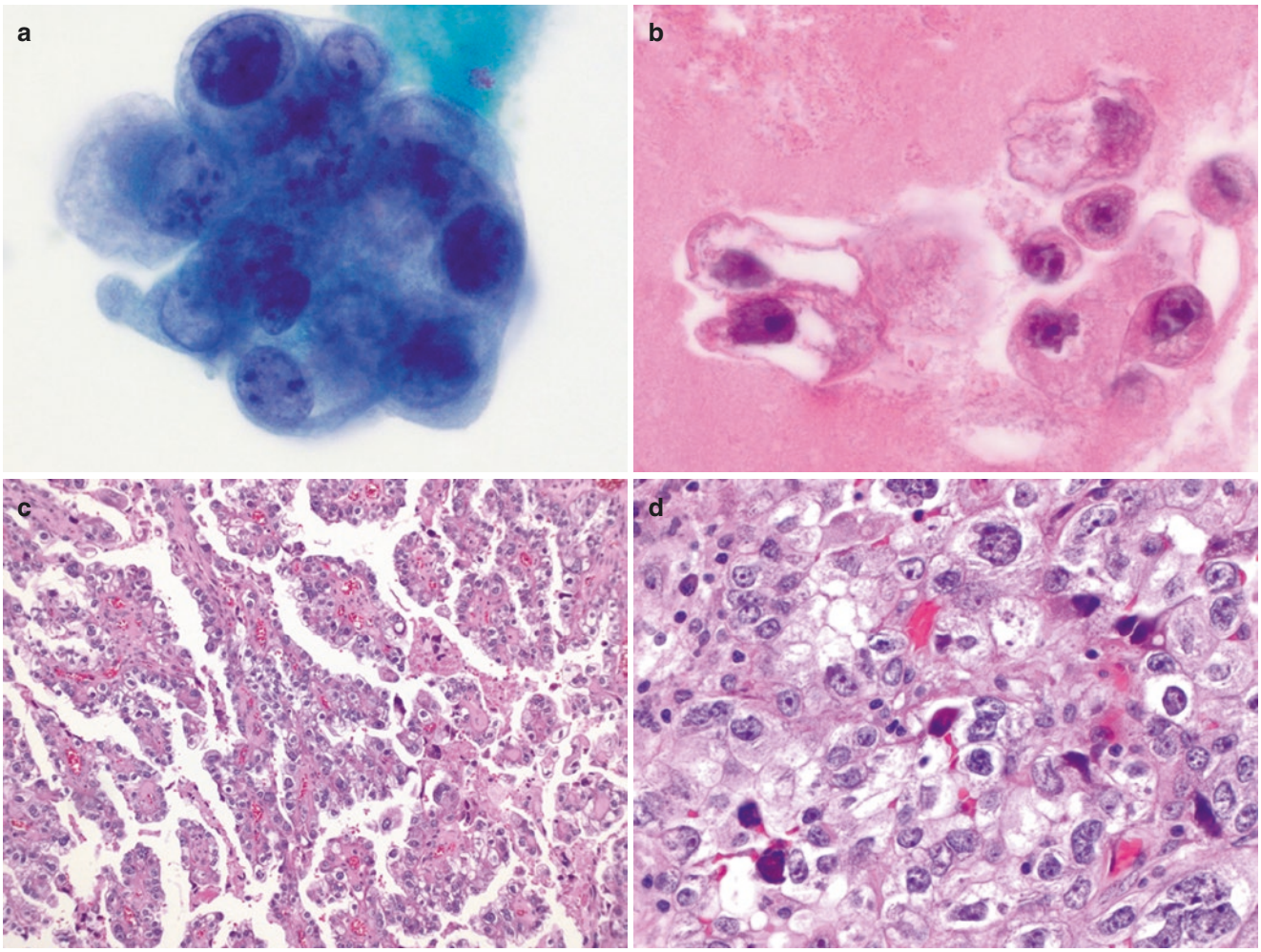

Fig. 11.9 Clear cell carcinoma of the ovary. Tumor cells have malignant nuclei and often exhibit abundant clear cytoplasm (**a**: ThinPrep, Papanicolaou stain; **b**: cell block, H&E stain). Resected tumor showed characteristic histologic features of clear cell carcinomas, namely malignant clear cells lining papillae in a "hobnail" appearance (**c**), and pleomorphic nuclei and abundant clear cytoplasm (**d**) (**c**, **d**: H&E stain)

26. What immunohistochemical markers are most helpful to subtype epithelial ovarian lesions? How should equivocal staining patterns be interpreted?

Subtyping of gynecologic malignancies has generally been reserved for surgical pathologists at the time of histologic resection, but advances in immunohistochemistry now provide cytopathologists with several tools for distinguishing gynecologic lesions (Table 11.5). Cytologic and histologic appearance and differential diagnosis should dictate the markers to be used. In the setting of equivocal staining patterns and a poorly differentiated gynecologic malignancy, a diagnosis of "high-grade Müllerian adenocarcinoma" can often be sufficient to guide further management.

27. What are the key cytologic and immunohistochemical features of germ cell tumors of the ovary?

Germ cell tumors of the ovary are analogous to those that arise in the testes. They most commonly present in women of reproductive age, and the vast majority of these are mature teratomas. However, care should be taken when examining specimens from pediatric patients, as malignant germ cell tumors are relatively much more common in this population.

Conceptually, there are three major categories of ovarian germ cell tumors, each of which have distinct cytomorphologic and immunohistochemical characteristics. If considering a germ cell tumor in the differential diagnosis, SALL-4 can be helpful first-line marker because it is positive in all three types of germ cell lesions. In cases of cytologically ambiguous germ cell tumors, the different types can often be distinguished by immunohistochemistry (Table 11.6).

- **Tumors with Embryonic Ectoderm, Mesoderm, and/or Endoderm Differentiation:** These tumors include mature and immature teratomas, the latter of which is malignant. Mature teratomas are not usually aspirated or biopsied because they frequently demonstrate characteristic ultrasonographic features. If aspirated, the most

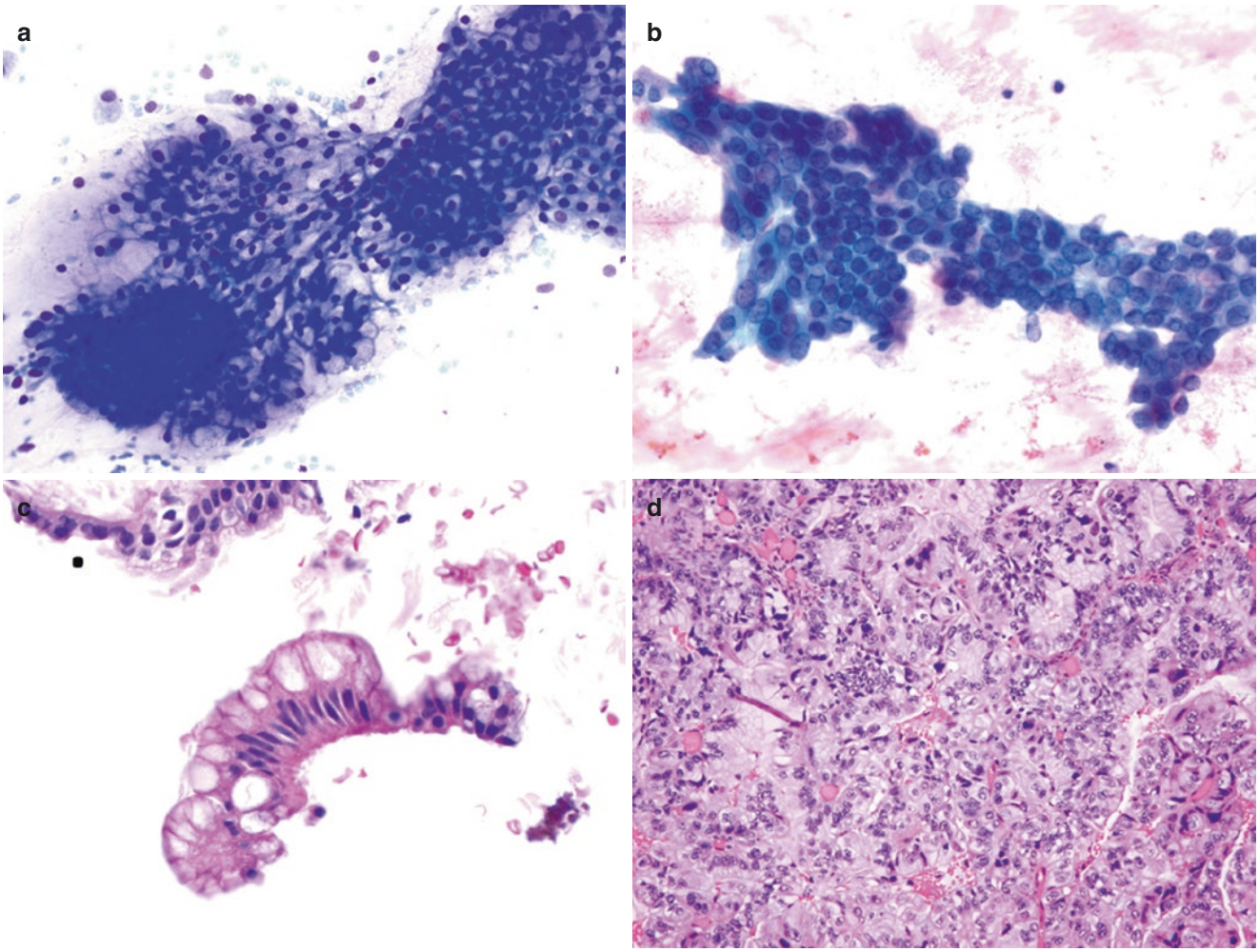

Fig. 11.10 Mucinous tumors of the ovary. FNA smear demonstrating sheets of benign-appearing mucinous epithelium (**a**: Diff-Quik stain; **b**: Papanicolaou stain) (**c**) Fragments of mucinous epithelium with goblet cells seen on a cell block, reminiscent of gastrointestinal epithelium. (**d**) At resection, mucinous tumors must be heavily sampled for focal areas of mucinous adenocarcinoma which are often missed on cytology (**c**, **d**: H&E stain)

Table 11.5 Summary of immunohistochemical staining of various epithelial ovarian tumors and mucinous metastases from gastrointestinal primaries

Ovarian tumor subtype	CK7	CK20	PAX-8	WT-1	CDX-2	SATB2	ER	PR	Napsin-A	AMACR	p53	p16
High-grade serous carcinoma	+	–	+	+	–	–	V	V	–	–	Mutant	Diffuse
Low-grade serous neoplasia	+	–	+	+	–	–	+	+	–	–	WT	-/F
Endometrioid type histology	+	–	+	–	–	–	+	+	–	–	V	V
Clear cell type histology	+	–	+	–	–	–	–	–	+	+	V	-/F
Mucinous (primary ovarian)	+	V	V	–	V	–	–	–	–	–	WT	-/F
Mucinous (metastatic)	–	+	–	–	+	+	–	–	–	–	V	-/F

Abbreviations and symbols: + positive, – negative, *V* variable, *WT* wild-type, *F* focal

Table 11.6 Immunohistochemical markers for malignant ovarian germ cell tumors

Malignant germ cell tumor	SALL-4	Pan-K	OCT-3/4	NANOG	c-Kit	AFP	hCG	GATA-3
Dysgerminoma (~50%)	+	Rare	+	+	+	–	–	–
Yolk sac tumor (~20%)	+	+	–	–	Rare	+	–	–
Embryonal carcinoma (3%)	+	+	+	+	–	–	–	–
Nongestational choriocarcinoma (1%)	+	+	–	–	–	–	+	+
Immature teratoma (20%)	+	–	+	–	–	–	–	–

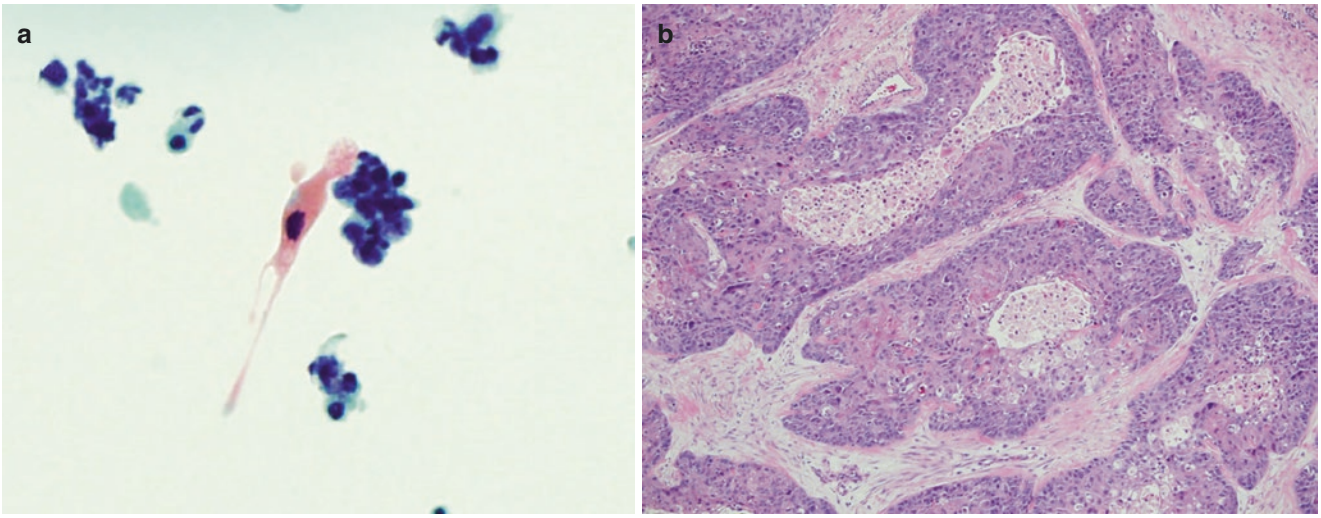

Fig. 11.11 Squamous cell carcinoma arising from mature teratoma. (**a**) Peritoneal washings from a patient showed dysplastic squamous cells in a background of reactive mesothelial cells (Papanicolaou stain). The differential diagnosis includes metastasis from a cervical lesion, but pap history and colposcopic findings may be negative. Immunohistochemistry with p16 may be helpful to exclude a cervical lesion. (**b**) Resection of an adnexal mass revealing squamous cell carcinoma arising from an ovarian mature teratoma (H&E stain)

common finding is anucleated squamous cells, which are indicative of ectodermal differentiation. Ectopic tissue from other organs can be present, including thyroid (struma ovarii), and, though uncommon, can undergo malignant transformation, leading to somatic-type malignancies such as primary ovarian thyroid carcinomas (positive for TTF-1 and PAX-8 by IHC) and squamous cell carcinomas (p63 positive by IHC, Fig. 11.11), which often portend a poor prognosis. All mature teratomas must be resected and evaluated for the presence of immature neuroectodermal elements, which are rare but diagnostic of malignancy.

- **Tumors that Express Transcription Factors of Pluripotency (i.e., OCT3/4 and NANOG):** This group of malignant germ cell tumors includes dysgerminomas (the ovarian equivalent of testicular seminomas) and embryonal carcinomas. Of the two, dysgerminomas are much more common, representing up to 5% of all ovarian malignancies. An accurate diagnosis of dysgerminoma is extremely important because these tumors respond well to therapy. Dysgerminomas often appear poorly differentiated cytologically, with large round nuclei, prominent nucleoli, and clear cytoplasm. Dysgerminomas will stain positive for the transcription factors associated with stem cell pluripotency, OCT-3/4 and NANOG. In fact, it is thought that dysgerminomas may be precursors of other germ cell tumors, a theory which may explain why 10% of germ cell tumors have mixed cell types. Embryonal carcinomas may stain positive for these markers as well. Cytologically, embryonal carcinoma will exhibit large round cells with irregular pleomorphic nuclei and multiple chromocenters. The two can often be distinguished by IHC, as dysgerminomas are typically keratin-negative and exhibit membranous staining for c-Kit (CD117).
- **Tumors with Extraembryonic Differentiation:** The germ cell tumors in this category include yolk sac tumor and nongestational choriocarcinoma. Both can appear poorly differentiated malignant epithelioid neoplasms, and the fact that they are keratin-positive can lead to a potential pitfall by rendering a diagnosis of carcinoma. Yolk sac tumors will be positive for alpha-fetoprotein and SALL-4. Choriocarcinomas will be positive for human chorionic gonadotropin (hCG), GATA-3, and SALL-4.

28. What are the key cytologic and immunohistochemical features of sex cord-stromal tumors of the ovary?

Sex cord-stromal tumors of the ovary are benign in 90% of cases. The class of benign sex cord-stromal tumors is comprised of fibromas (which derived from ovarian stromal fibroblasts), thecomas (derived from hormone-secreting ovarian stromal cells), or mixed tumors with features of both fibromas and thecomas. Aspirates of these tumors are generally hypocellular, because they are often solid tumors with abundant intracellular collagen. Fibromas are composed of spindle-shaped cells with an unsurprising fibroblast-like appearance. Pure thecomas are rare, but thecomatous cells generally have a monomorphic appearance with clear cytoplasm and varying degrees of vacuolization.

Sex cord-stromal tumors of the ovary with malignant potential include adult granulosa cell tumor, juvenile granulosa cell tumor, Sertoli-Leydig cell tumor, and steroid cell tumors. Not all sex cord tumors of the ovary are easily cate-

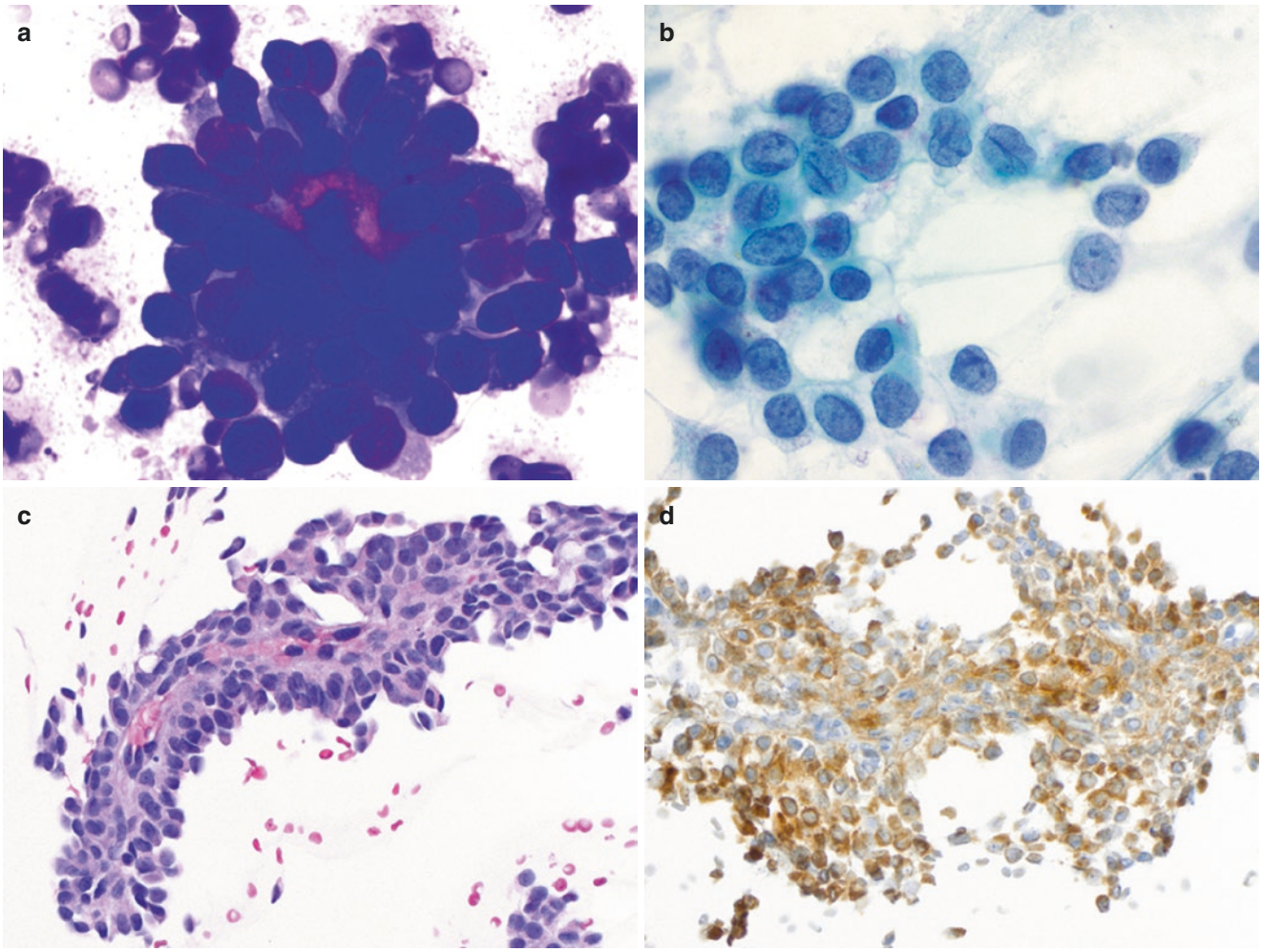

Fig. 11.12 Adult granulosa cell tumor (AGCT). (**a**) Diff-Quik stain of an AGCT illustrating a pseudofollicular arrangement (Call-Exner body). (**b**) The nuclear grooves and irregularities ("coffee bean nuclei") of AGCT are best visualized on alcohol-fixed preparations (Papanicolaou stain). (**c**) Cell block of AGCT, demonstrating a papillary-like architecture (H&E stain). (**d**) Inhibin IHC is commonly used to diagnose AGCT, which will demonstrate cytoplasmic positivity

gorized, sometimes prompting a diagnosis of "unclassified sex cord-stromal tumor."

Adult granulosa cell tumors (AGCT) are typically unilateral and confined to the ovary, but patients can present with ruptured tumors or peritoneal spread. These tumors are predominantly solid, composed of monomorphic neoplastic cells with scant cytoplasm that can appear singly, as naked nuclei, in loose clusters, in cords, or in a pseudofollicular pattern (Fig. 11.12). This latter architecture, termed Call-Exner bodies, describes a pattern of granulosa cells arranged around small globules of eosinophilic hyaline. Other helpful cytologic features of granulosa cells include prominent nuclear grooves and nuclear membrane irregularity. The distinction between AGCT and follicular cysts can be particularly challenging, as immunohistochemistry is unlikely to help, but the presence of predominantly normal granulosa cells can be helpful.

As the name suggests, juvenile granulosa cell tumors (JGCT) are more commonly seen in young patients. There are a few features which distinguish the juvenile from the adult variants. Foremost, there are genetic differences between the two because the vast majority of AGCTs harbor mutations of the transcription factor, *FOXL2*, which is wild-type in 90% of JGCTs [47]. From a cytologic perspective, the tumor cells of JGCT lack nuclear grooves and Call-Exner bodies, and the tumor nuclei appear round with fine chromatin and small chromocenters.

Immunohistochemistry can be very helpful in distinguishing sex cord-stromal tumors from other ovarian lesions. WT-1 and SF-1 are considered pan-markers of sex cord-stromal tumors [48]. Other markers, such as inhibin, calretinin, and CD99, are variably expressed in benign sex cord-stromal tumors, although they are more often positive in the malignant entities.

29. What are the key cytologic and immunohistochemical characteristics of small cell carcinomas of the ovary?

Small cell carcinomas of the ovary are malignant tumors composed of small round blue undifferentiated cells and are divided into two entities:

- **Small Cell Carcinoma of the Ovary, *Hypercalcemic Type*:** A poorly differentiated epithelial tumor that is associated with paraneoplastic hypercalcemia in 62% of cases. These tumors typically occur in younger patients (mean 24 years of age) and have a poor overall survival rate. Tumors will express CD56, synaptophysin, and, occasionally, parathyroid-related hormone. Inactivating mutations of the chromatin remodeling enzyme, SMARCA4, are often found in these tumors [49], and thus immunohistochemistry for SMARCA4 loss can particularly be helpful in the diagnosis, which distinguishes it from its pulmonary type counterpart.
- **Small Cell Carcinoma of the Ovary, *Pulmonary Type*:** A poorly differentiated tumor that expresses neuroendocrine markers analogous to small cell carcinoma of the lung. The primary differential diagnosis includes hypercalcemic type primary ovarian small cell carcinoma and metastasis from a lung primary. The presence of a lung mass and TTF-1 positivity by IHC favors a metastatic lesion.

30. What are the key features of metastatic tumors of the ovary?

Metastatic lesions to the ovary are common and account for almost 10% of malignant ovarian neoplasms found in women undergoing surgery for an adnexal mass. Common features of metastasis to the ovary include bilateral ovarian involvement, surface involvement, a nodular pattern of spread, small size (<10 cm), and history of a known nonovarian primary malignancy. In contrast, primary ovarian tumors are typically unilateral, large (>10 cm). Ovarian involvement commonly presents with metastases from colorectal (37%), breast (12%), gastric (9%), appendiceal (9%), pancreas (6%), and lung (2%) primaries.

Peritoneal Washings

1. What is the purpose of peritoneal washings?

The primary purpose of peritoneal washing cytology is to identify metastatic disease in the peritoneum that is not grossly visible, typically at the time of staging laparoscopy or resection. Peritoneal washings are obtained during benign gynecologic procedures to help exclude occult disease. In the setting of known metastatic disease, peritoneal washings can be used to monitor treatment response.

Washings are particularly important for gynecologic oncologists because cytologic evaluation is part of the staging system for fallopian tube and ovarian cancers [50]. Historically, peritoneal washings were evaluated in the staging of endometrial cancers (indicating stage IIIA disease), but the International Federation of Gynecology and Obstetrics (FIGO) revised staging criteria in 2009, and washing cytology was removed from the staging criteria [51].

2. What are the prognostic implications of positive peritoneal washings in gynecologic malignancies?

For endometrial cancers (stage I to IIIa), peritoneal washing cytology is an independent predictor of disease recurrence and mortality, and, in advanced stage patients, metastasis to the adnexa or uterine serosa does not seem to confer a worse prognosis than positive cytology alone [52]. It has been discovered that laparoscopically assisted vaginal hysterectomies have a higher incidence of positive peritoneal cytology compared to total abdominal hysterectomy, possibly due to retrograde dislocation of cancer cells during manipulation of the uterus [53], but the clinical significance of this appears to be minimal [54].

For cancers of the fallopian tube and ovary, tumors associated with positive peritoneal washings or ascites are classified as FIGO stage IC3, if they are otherwise confined to the adnexa and the washings are not associated with surgical spill intraoperatively (IC1), capsule rupture prior to surgery (IC2), or tumor on the ovarian surface (also IC2) [55]. Of note, the American Joint Committee on Cancer (AJCC) TNM staging does not make this distinction, and these three sub-stages are considered together simply as T1c. For invasive epithelial ovarian cancer, the 5-year survival for patients with stage IC is 81%, compared to 92% for IA and 14% for stage IV malignancies [56].

3. How often do peritoneal washings change the surgical staging in patients with gynecologic cancers?

In reality, surgical staging may not change very often, and this may be one of the reasons that washing status was eliminated from the FIGO staging criteria for endometrial cancer. A positive peritoneal washing upstages only 4.5% of patients and does not appear to affect outcomes [57].

For ovarian cancer, it has been estimated that peritoneal washings will upstage as many as 25% of patients with low stage disease [58], but it is important to note that the detection rate of peritoneal washings in otherwise stage IA or IB patients is heavily dependent on tumor subtype, with serous carcinomas more often positive than other variants [59]. FIGO stage IC (TNM T1c) accounts for 18.7% of all ovarian tumors [56].

4. Does ovarian cyst rupture during surgery lead to worse prognosis in the absence of surface involvement or positive ascites/washings?

This remains a controversial issue, with some studies finding a higher risk of recurrence and others not. Ovarian cyst rupture is always avoided if possible during primary resection of tumors confined to the adnexa because multivariate analysis has shown that capsule rupture and positive peritoneal washings are independent predictors of poor prognosis [55].

5. Are peritoneal washings obtained in any nongynecologic surgeries?

Although not part of the TNM staging of any nongynecologic malignancies, positive washings are often obtained during resection procedures because they are associated with poor prognosis in abdominal malignancies:

- In patients with gastric adenocarcinomas, positive washings are associated with advanced stage and poor overall survival [60].
- In patients undergoing surgery for colorectal cancer, positive peritoneal washings had a significantly higher rate of local recurrence and peritoneal carcinomatosis than those with negative washings [61], and long-term follow-up has revealed that the 10-year survival rate for patients with positive cytology is less than those with negative washings [62].
- In patients with pancreatic cancer, there is a significant correlation between positive peritoneal cytology and the presence of peritoneal metastases [63], and survival is typically worse than patients with negative cytology [64].

6. How accurate is peritoneal washing cytology versus ascites? If a peritoneal biopsy is positive for malignant cells, do peritoneal washings provide any additional information?

There are a number of important concerns about the accuracy of peritoneal washings. First, many patients with metastases to the peritoneum will have negative washings, with up to ~50% of patient's having false-negative cytology [65]. However, as stated above, the detection rate (and therefore the sensitivity and specificity) is highly dependent on tumor subtype [59]. For instance, in cases of low-grade serous neoplasia, the sensitivity is relatively high and strongly correlates with ovarian surface involvement and peritoneal implants [66].

Evaluation of ascites fluid has a false-negative rate that is about 6%, much less than that of peritoneal washings [65]. If histologic biopsy confirms peritoneal involvement, peritoneal washings provide no additional information, and the patient will be staged based on the results of the biopsy.

7. What common conditions lead to false-positive washings?

False-positive peritoneal washings occur in less than 5% of case [67], which can result in the following conditions:

- Mesothelial proliferation with psammomatous calcifications [68]
- Endometriosis, particularly with eosinophilic metaplasia [69]
- Endosalpingiosis [70]
- Ectopic pancreas [71]

The presence of Müllerian epithelium associated with psammomatous calcifications should prompt the cytologist to render an atypical diagnosis (Fig. 11.13).

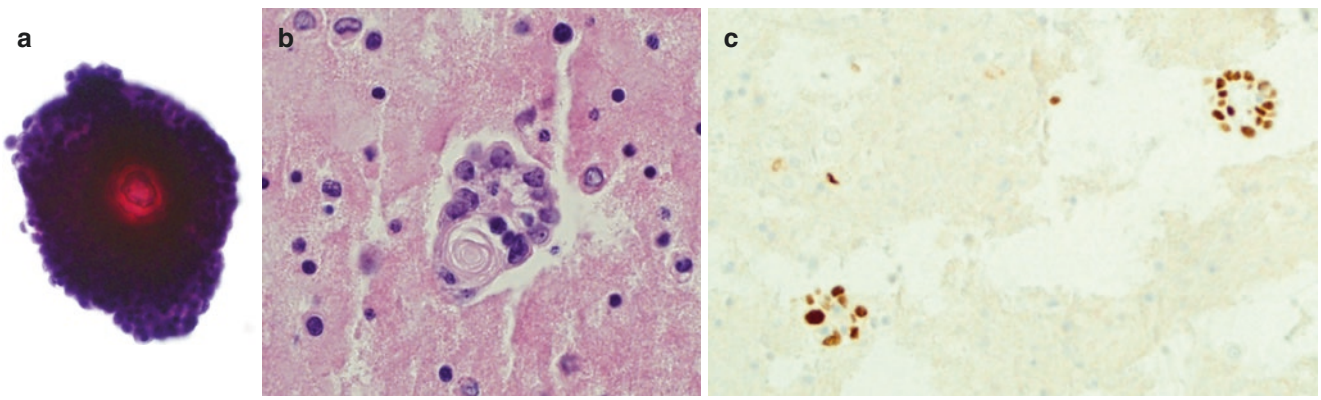

Fig. 11.13 Benign-appearing Müllerian proliferations associated with psammomatous calcifications. (**a**) Diff-Quik stain demonstrating a three-dimensional cluster of small, benign-appearing cells associated with a psammoma body. The differential diagnosis includes a mesothelial proliferation versus a benign Müllerian inclusion (such as endometriosis or endosalpingiosis) versus an implant of low-grade serous neoplasia. (**b**) Cell block with H&E stain demonstrating epithelium associated with a concentrically laminated calcification. Unless stroma is present, it is not possible to tell if the cells are derived from an invasive or a noninvasive implant. (**c**) Immunohistochemistry for PAX-8 will exhibit strong nuclear positivity in Müllerian epithelium, which is usually negative in mesothelial cells

8. Is there any reason to segregate washings from different peritoneal sites?
Peritoneal washings should be obtained from different peritoneal locations, which should be combined into a single specimen. There appears to be no benefit to segregating samples [72].

9. What are the adequacy criteria for a peritoneal washing specimen?
Strict adequacy criteria have not been established for peritoneal washings, but the presence of benign mesothelial cells should be identified before considering a specimen adequate. If malignant cells are present, the specimen should also be considered adequate [67].

10. How useful are atypical and suspicious interpretations of peritoneal cytology?
In general, peritoneal washings that are interpreted as "atypical" or "suspicious" are not useful to clinicians, and anything less than a malignant diagnosis is considered as a negative result [67].

Case Presentations

Case 1
Learning Objectives:

1. Review the cytology of mucinous tumors of the ovary.
2. Generate a differential diagnosis for mucinous tumors of the ovary.
3. Understand how IHC can differentiate primary ovarian from metastatic mucinous lesions in the ovary.

Case History:

- A 58-year-old female presents with abdominal distention and an elevated CA-125. CT reveals a 22-cm multiloculated cystic pelvic mass. The ovaries are not well visualized on imaging.

Specimen Source:

- U/S-guided FNA of a pelvic mass

Cytologic Findings:

- Abundant mucin admixed with inflammatory cells (Fig. 11.14a).
- Small group of mucinous cells without significant cytologic atypia (Fig. 11.14b).
- Cell block demonstrating strips of mucinous epithelium and stroma (Fig. 11.14c).
- Low-grade components are more likely to be found in primary ovarian mucinous tumors than metastases.

Differential Diagnosis:

- Mucinous cystadenoma of the ovary
- Mucinous borderline tumor of the ovary
- Mucinous adenocarcinoma of the ovary
- Metastatic mucinous adenocarcinoma

IHC and Other Ancillary Studies:

- Definitive subtyping requires resection and histologic evaluation for invasion.
- CK7 and PAX-8 positivity would favor an ovarian primary.
- CK20 and CDX-2 positivity would **not** exclude an ovarian primary.
- SATB2 would strongly favor metastasis from an appendiceal or a colorectal primary.

Final Cytologic Diagnosis:
Mucinous cystic neoplasm
Take-Home Messages:

1. Mucinous tumors of the ovary can be benign or malignant.
2. Mucinous tumors of the ovary require resection and histologic evaluation to correctly subtype.
3. Immunohistochemistry is often not definitive but can be helpful to favor an ovarian primary.

References: [43–46]

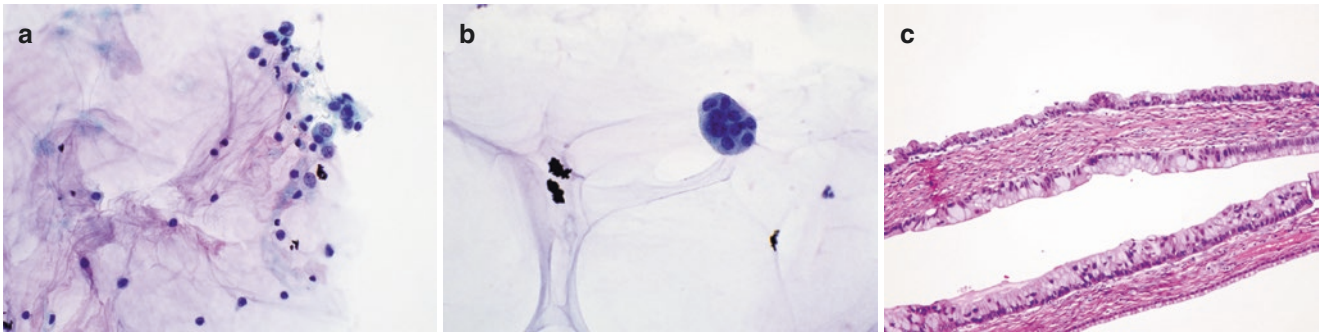

Fig. 11.14 Case 1: Pelvic mass, FNA. (**a**, **b**) Images of alcohol-fixed smear (Papanicolaou stain). (**c**) Image of cell block preparation (H&E stain)

Case 2

Learning Objectives:

1. Review the cytology of tubo-ovarian high-grade serous carcinoma.
2. Generate a differential diagnosis for tubo-ovarian high-grade serous carcinoma.
3. Review the immunohistochemical profile of tubo-ovarian high-grade serous carcinoma.

Case History:

- A 78-year-old female presents with a 5-cm solid and cystic ovarian mass.

Specimen Source:

- U/S-guided FNA of the ovarian mass

Cytologic Findings:

- Hypercellular aspirate with papillary structures (Fig. 11.15).
- Cells can also appear singly or in crowded clusters.
- Cells will exhibit a high N:C ratio, nuclear irregularities, and mitotic figures.

Differential Diagnosis:

- Tubo-ovarian high-grade serous carcinoma
- Ovarian endometrioid adenocarcinoma
- Clear cell carcinoma of the ovary
- Metastatic adenocarcinoma

IHC and Other Ancillary Studies:

- Gynecologic malignancies can often be distinguished from metastatic lesions and will often be positive for CK7 and PAX-8.
- Positivity for WT-1 and p16 may favor an ovarian primary, but uterine carcinomas can present with this immunophenotype as well.
- High-grade serous carcinomas of the ovary and uterus often demonstrates a p53-mutant phenotype, in contrast to low-grade endometrioid adenocarcinomas and clear cell carcinomas of the ovary.

Final Cytologic Diagnosis:
High-grade serous carcinoma
Take-Home Messages:

1. High-grade serous carcinoma will exhibit malignant cytologic features and papillary architecture. In contrast, benign and low-grade serous lesions will not exhibit this degree of cytologic atypia.
2. The primary site of high-grade serous carcinoma may be the ovary, the fallopian tubes, or the uterus, and it is not possible to make this distinction by cytology.
3. A diagnosis of "high-grade Müllerian adenocarcinoma" is often sufficient to guide management.

References: [32, 73, 74]

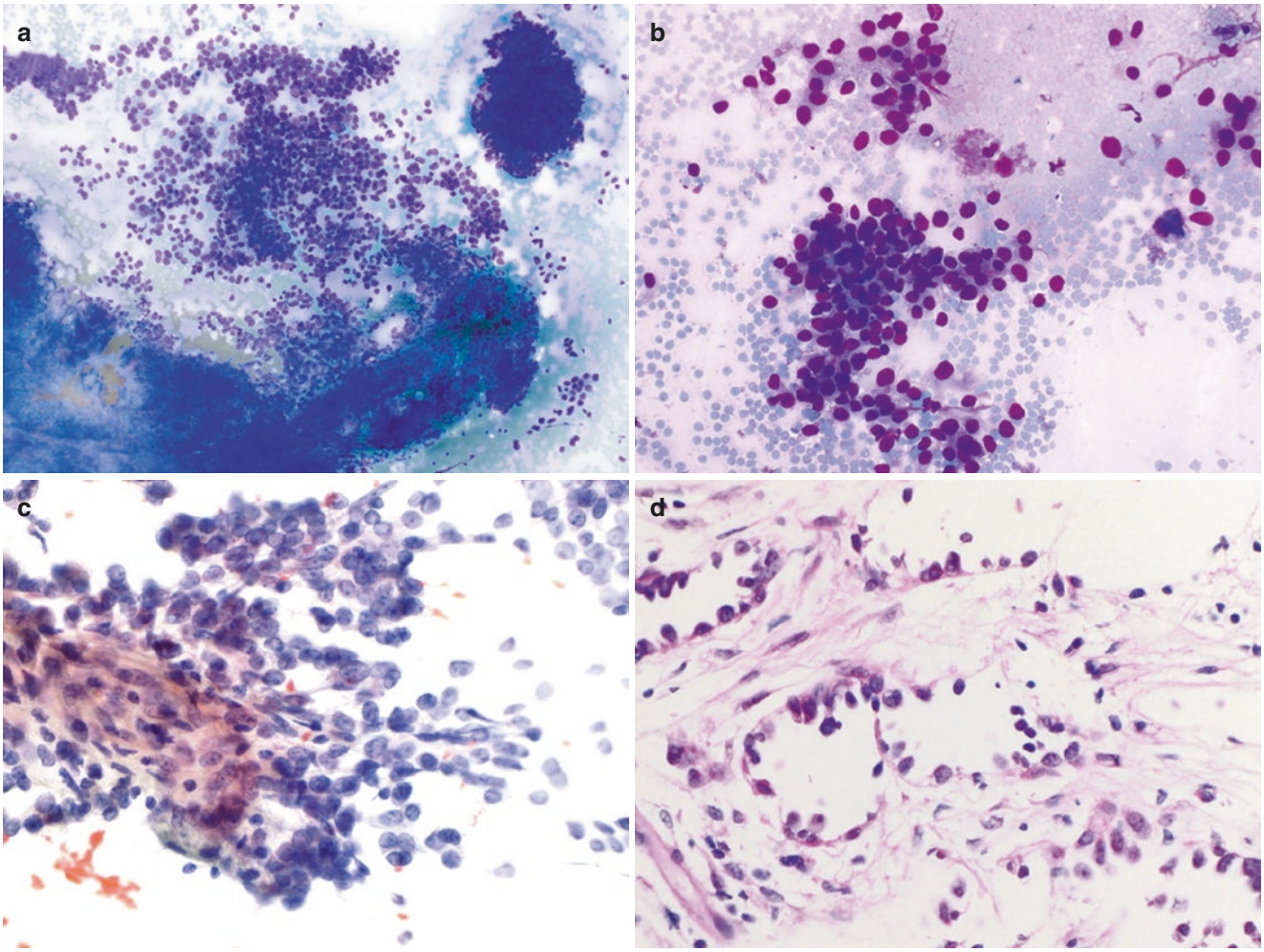

Fig. 11.15 Case 2: Ovarian mass, FNA. (**a**, **b**) Photomicrograph of air-dried smear (Diff-Quik stain). (**c**) Photomicrograph of alcohol-fixed smear (Papanicolaou stain). (**d**) Material from cell block (H&E stain)

Case 3

Learning Objectives:

1. Understand why peritoneal washings are obtained during gynecologic procedures.
2. Generate a differential diagnosis for endometrioid adenocarcinoma.
3. Understand the immunohistochemical profile of tubo-ovarian high-grade serous carcinoma.

Case History:

- A 40-year-old obese female with a history of cervical neoplasia presents with an adnexal mass and abnormal uterine bleeding. Endometrial biopsy demonstrates small fragments of adenocarcinoma, not otherwise specified. Hysterectomy and bilateral salpingo-oophorectomy are performed.

Specimen Source:

- Peritoneal washing

Cytologic Findings:

- Clusters of malignant glandular cells with increased N:C ratio, enlarged nuclei, and prominent nucleoli (Fig. 11.16a).
- Keratinized dysplastic cells are present in the washings (Fig. 11.16b).
- Malignant cells appear in the cell block, associated with numerous neutrophils (Fig. 11.16c). The source of keratinized cells is identified as squamous metaplasia on resection (Fig. 11.16d).

Differential Diagnosis:

- Endometrial versus ovarian endometrioid adenocarcinoma
- Serous carcinoma
- Endocervical adenocarcinoma

IHC and Other Ancillary Studies:

- Endometrioid adenocarcinomas are typically p53 wild-type, which distinguishes them from serous carcinomas.
- Immunohistochemistry for p16 will be negative or focal in endometrial adenocarcinomas, in contrast to HPV-related endocervical adenocarcinomas, which will be strong and diffusely p16 positive.

Final Cytologic Diagnosis:
Endometrioid adenocarcinoma

Take-Home Messages:

1. Peritoneal washings provide important prognostic information for ovarian and endometrial carcinomas, and washing status is a staging component of ovarian but not uterine cancers.
2. Endometrioid adenocarcinomas will have wild-type p53 and can exhibit squamous differentiation, which distinguishes them from gynecologic serous carcinomas.
3. Endocervical adenocarcinomas are often in the differential diagnosis for young patients who are HPV-positive or have a history of cervical dysplasia.

References: [75, 76]

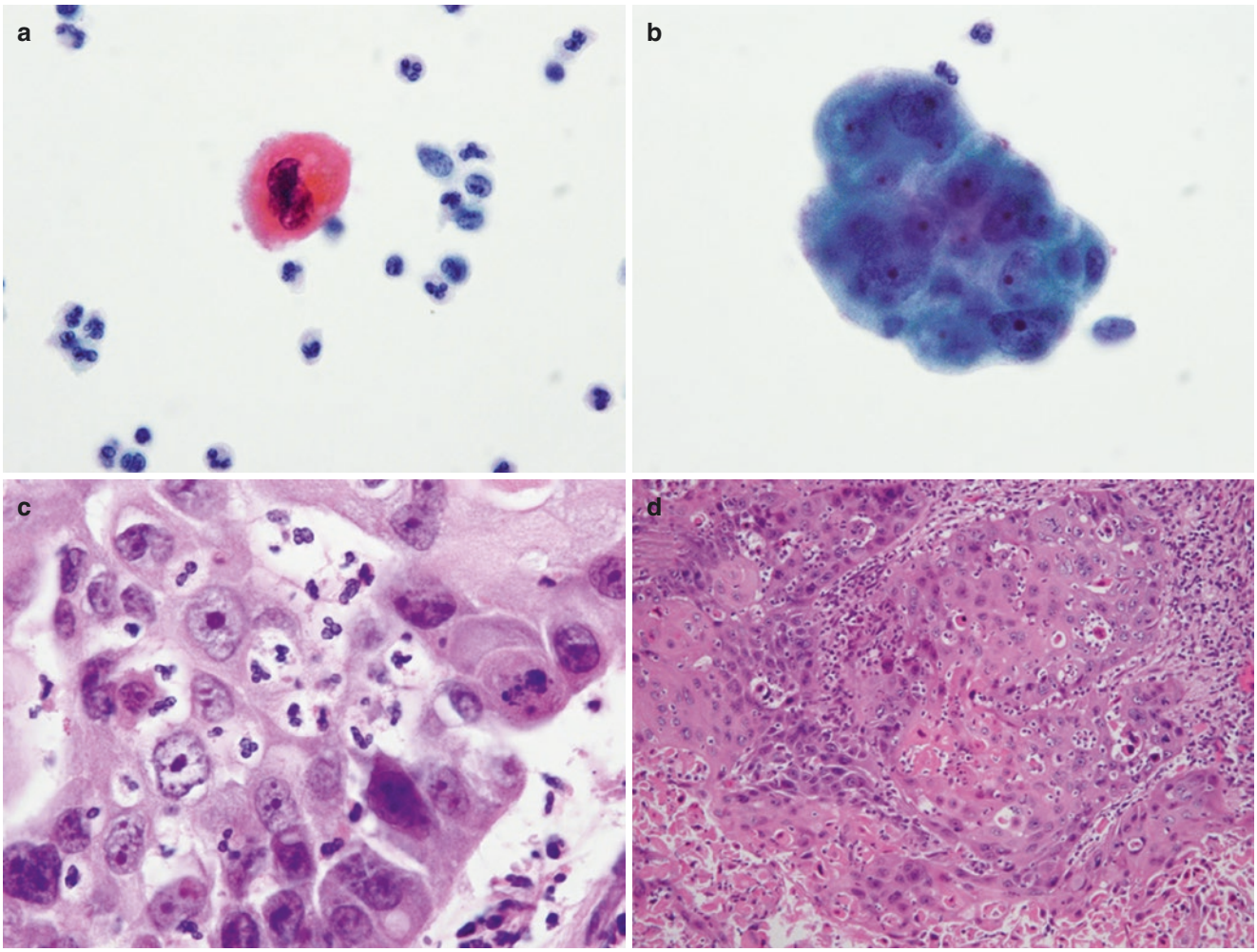

Fig. 11.16 Case 3: Cytology of pelvic washings obtained during hysterectomy. (**a**, **b**) Images taken from ThinPrep slide (Papanicolaou stain). (**c**) High-power image from cell block (H&E stain). (**d**) Resection of uterine tumor (H&E stain)

References

1. Kjellgren O, Angström T, Bergman F, Wiklund DE. Fine-needle aspiration biopsy in diagnosis, and classification of ovarian carcinoma. Cancer. 1971;28(4):967–76. http://www.ncbi.nlm.nih.gov/pubmed/5111748. Accessed 11 Feb 2018.
2. Angstrom T, Kjellgren O, Bergman F. The cytologic diagnosis of ovarian tumors by means of aspiration biopsy. Acta Cytol. 1972;16(4):336–41. http://www.ncbi.nlm.nih.gov/pubmed/4504235. Accessed 11 Feb 2018.
3. Khaw KT, Walker WJ. Ultrasound guided fine needle aspiration of ovarian cysts: diagnosis and treatment in pregnant and non-pregnant women. Clin Radiol. 1990;41(2):105–8. https://doi.org/10.1016/S0009-9260(05)80139-4.
4. Ateş O, Karakaya E, Hakgüder G, Olguner M, Seçil M, Akgür FM. Laparoscopic excision of a giant ovarian cyst after ultrasound-guided drainage. J Pediatr Surg. 2006;41(10):e9–e11. https://doi.org/10.1016/J.JPEDSURG.2006.06.023.
5. Oleszczuk J, Leszczyńska-Gorzelak B, Kamiński K, Baranowski W. Evaluation of ovarian cysts by means of transabdominal fine needle aspiration or laparotomy in pregnant women. Ginekol Pol. 2002;73(4):367–370. http://www.ncbi.nlm.nih.gov/pubmed/12152287. Accessed 11 Feb 2018.
6. CRESPIGNY LC, ROBINSON HP, DAVOREN RAM, FORTUNE D. The "simple" ovarian cyst: aspirate or operate? BJOG An Int J Obstet Gynaecol. 1989;96(9):1035–9. https://doi.org/10.1111/j.1471-0528.1989.tb03377.x.
7. Trimbos JB, Hacker NF. The case against aspirating ovarian cysts. Cancer. 1993;72(3):828–31. https://doi.org/10.1002/1097-0142(19930801)72:3<828::AID-CNCR2820720331>3.0.CO;2-T.

8. Duke D, Colville J, Keeling A, Broe D, Fotheringham T, Lee MJ. Transvaginal aspiration of ovarian cysts: long-term follow-up. Cardiovasc Intervent Radiol. 2006;29(3):401–5. https://doi.org/10.1007/s00270-005-0167-0.
9. Koutlaki N, Nikas I, Dimitraki M, et al. Transvaginal aspiration of ovarian cysts: our experience over 121 cases. Minim Invasive Ther Allied Technol. 2011;20(3):155–9. https://doi.org/10.3109/13645706.2010.532220.
10. Nikolaou M, Adonakis G, Zyli P, et al. Transvaginal ultrasound-guided aspiration of benign ovarian cysts. J Obstet Gynaecol (Lahore). 2014;34(4):332–5. https://doi.org/10.3109/01443615.2013.874406.
11. Higgins RV, Matkins JF, Marroum M-C. Comparison of fine-needle aspiration cytologic findings of ovarian cysts with ovarian histologic findings. Am J Obstet Gynecol. 1999;180(3):550–3. https://doi.org/10.1016/S0002-9378(99)70252-8.
12. Bonilla-Musoles F, Ballester MJ, Simon C, Serra V, Raga F. Is avoidance of surgery possible in patients with perimenopausal ovarian tumors using transvaginal ultrasound and duplex color Doppler sonography? J Ultrasound Med. 1993;12(1):33–9. https://doi.org/10.7863/jum.1993.12.1.33.
13. Geier GR, Strecker JR. Aspiration cytology and E2 content in ovarian tumors. Acta Cytol. 1981;25(4):400–6. http://www.ncbi.nlm.nih.gov/pubmed/6945006. Accessed 11 Feb 2018.
14. Moran O, Menczer J, Ben-Baruch G, Lipitz S, Goor E. Cytologic examination of ovarian cyst fluid for the distinction between benign and malignant tumors. Obstet Gynecol. 1993;82(3):444–6. http://www.ncbi.nlm.nih.gov/pubmed/8355950. Accessed 11 Feb 2018.
15. Zhou AG, Levinson KL, Rosenthal DL, VandenBussche CJ. Performance of ovarian cyst fluid fine-needle aspiration cytology. Cancer Cytopathol. 2018;126(2):112–21. https://doi.org/10.1002/cncy.21911.
16. Sevelda P, Dittrich C, Salzer H. Prognostic value of the rupture of the capsule in stage I epithelial ovarian carcinoma. Gynecol Oncol. 1989;35(3):321–2. https://doi.org/10.1016/0090-8258(89)90071-1.
17. Dembo AJ, Davy M, Stenwig AE, Berle EJ, Bush RS, Kjorstad K. Prognostic factors in patients with stage I epithelial ovarian cancer. Obstet Gynecol. 1990;75(2):263–73. http://www.ncbi.nlm.nih.gov/pubmed/2300355. Accessed 11 Feb 2018.
18. Trimbos JB, Schueler JA, van Lent M, Hermans J, Fleuren GJ. Reasons for incomplete surgical staging in early ovarian carcinoma. Gynecol Oncol. 1990;37(3):374–7. https://doi.org/10.1016/0090-8258(90)90370-Z.
19. Sjovall K, Nilsson B, Einhorn N. Different types of rupture of the tumor capsule and the impact on survival in early ovarian carcinoma. Int J Gynecol Cancer. 1994;4(5):333–6. https://doi.org/10.1046/j.1525-1438.1994.04050333.x.
20. Zanetta G, Trio D, Lissoni A, et al. Early and short-term complications after US-guided puncture of gynecologic lesions: evaluation after 1,000 consecutive cases. Radiology. 1993;189(1):161–4. https://doi.org/10.1148/radiology.189.1.8372188.
21. Livraghi T, Damascelli B, Lombardi C, Spagnoli I. Risk in fine-needle abdominal biopsy. J Clin Ultrasound. 1983;11(2):77–81. https://doi.org/10.1002/jcu.1870110205.
22. Spanos WJ. Preoperative hormonal therapy of cystic adnexal masses. Am J Obstet Gynecol. 1973;116(4):551–6. https://doi.org/10.1016/0002-9378(73)90913-7.
23. Brown DL, Doubilet PM, Miller FH, et al. Benign and malignant ovarian masses: selection of the most discriminating gray-scale and Doppler sonographic features. Radiology. 1998;208(1):103–10. https://doi.org/10.1148/radiology.208.1.9646799.
24. Mulvany N, Ostör A, Teng G. Evaluation of estradiol in aspirated ovarian cystic lesions. Acta Cytol. 1995;39(4):663–8. http://www.ncbi.nlm.nih.gov/pubmed/7631540. Accessed 11 Feb 2018.
25. Allias F, Chanoz J, Blache G, Thivolet-Bejui F, Vancina S. Value of ultrasound-guided fine-needle aspiration in the management of ovarian and paraovarian cysts. Diagn Cytopathol. 2000;22(2):70–80. https://doi.org/10.1002/(SICI)1097-0339(200002)22:2<70::AID-DC3>3.0.CO;2-J.
26. Stanley MW, Horwitz CA, Frable WJ. Cellular follicular cyst of the ovary: fluid cytology mimicking malignancy. Diagn Cytopathol. 1991;7(1):48–52. https://doi.org/10.1002/dc.2840070113.
27. Nuñez C, Diaz JI, Selvaggi S. Ovarian follicular cysts: a potential source of false positive diagnoses in ovarian cytology. Diagn Cytopathol. 1992;8(5):532–7. https://doi.org/10.1002/dc.2840080515.
28. Wojcik EM, Selvaggi SM. Fine-needle aspiration cytology of cystic ovarian lesions. Diagn Cytopathol. 1994;11(1):9–14. https://doi.org/10.1002/dc.2840110104.
29. Zaloudek CJ, Garg K. Tumors of the female genital tract: ovary, fallopian tube, and broad and round ligaments. In: Fletcher CDM, editor. Diagnostic histopathology of tumors 2. 4th ed. Philadelphia, PA: Elsevier; 2013. p. 658–761.
30. Nucci M, Oliva E. Diagnostic pathology: gynecological. 1st ed. Philadelphia, PA: Amirsys; 2014.
31. Kurman R, Carcangiu M, Herrington C, Young R, editors. WHO classification of tumours of female reproductive organs. 4th ed. Lyon: IARC Press; 2014.
32. Malpica A, Deavers MT, Lu K, et al. Grading ovarian serous carcinoma using a two-tier system. Am J Surg Pathol. 2004;28(4):496–504. http://www.ncbi.nlm.nih.gov/pubmed/15087669. Accessed 21 Feb 2018.
33. Neely Larry Copeland EJ, Gershenson DM, Taylor Wharton J, Sneige N, Edwards CL, Rutledge FN. Microscopic disease at second-look laparotomy in advanced ovarian cancer. https://onlinelibrary.wiley.com/doi/pdf/10.1002/1097-0142%2819850115%2955%3A2%3C472%3A%3AAID-CNCR2820550231%3E3.0.CO%3B2-A. Accessed 18 Feb 2018.
34. Cancer Genome Atlas Research Network. Integrated genomic analyses of ovarian carcinoma. Nature. 2011;474(7353):609–15. https://doi.org/10.1038/nature10166.
35. Cole AJ, Dwight T, Gill AJ, et al. Assessing mutant p53 in primary high-grade serous ovarian cancer using immunohistochemistry and massively parallel sequencing. Sci Rep. 2016;6(1):26191. https://doi.org/10.1038/srep26191.
36. Singer G, Stöhr R, Cope L, et al. Patterns of p53 mutations separate ovarian serous borderline tumors and low- and high-grade carcinomas and provide support for a new model of ovarian carcinogenesis. Am J Surg Pathol. 2005;29(2):218–24. https://doi.org/10.1097/01.pas.0000146025.91953.8d.
37. Okuda T, Otsuka J, Sekizawa A, et al. p53 mutations and overexpression affect prognosis of ovarian endometrioid cancer but not clear cell cancer. Gynecol Oncol. 2003;88(3):318–25. https://doi.org/10.1016/S0090-8258(02)00149-X.
38. Ho ES, Lai CR, Hsieh YT, et al. p53 mutation is infrequent in clear cell carcinoma of the ovary. Gynecol Oncol. 2001;80(2):189–93. https://doi.org/10.1006/gyno.2000.6025.
39. Tashiro H, Isacson C, Levine R, Kurman RJ, Cho KR, Hedrick L. p53 gene mutations are common in uterine serous carcinoma and occur early in their pathogenesis. Am J Pathol. 1997;150(1):177–85. http://www.ncbi.nlm.nih.gov/pubmed/9006334. Accessed 18 Feb 2018.
40. Lax SF. Molecular genetic pathways in various types of endometrial carcinoma: from a phenotypical to a molecular-based classification. Virchows Arch. 2004;444(3):213–23. https://doi.org/10.1007/s00428-003-0947-3.
41. Fadare O, Gwin K, Desouki MM, et al. The clinicopathologic significance of p53 and BAF-250a (ARID1A) expression in clear cell carcinoma of the endometrium. Mod Pathol. 2013;26(8):1101–10. https://doi.org/10.1038/modpathol.2013.35.

42. DeLair D, Oliva E, Köbel M, Macias A, Gilks CB, Soslow RA. Morphologic spectrum of immunohistochemically characterized clear cell carcinoma of the ovary. Am J Surg Pathol. 2011;35(1):36–44. https://doi.org/10.1097/PAS.0b013e3181ff400e.
43. Lee KR, Scully RE. Mucinous tumors of the ovary: a clinicopathologic study of 196 borderline tumors (of intestinal type) and carcinomas, including an evaluation of 11 cases with "pseudomyxoma peritonei". Am J Surg Pathol. 2000;24(11):1447–64. http://www.ncbi.nlm.nih.gov/pubmed/11075847. Accessed 21 Feb 2018.
44. Lee KR, Young RH. The distinction between primary and metastatic mucinous carcinomas of the ovary: gross and histologic findings in 50 cases. Am J Surg Pathol. 2003;27(3):281–92. http://www.ncbi.nlm.nih.gov/pubmed/12604884. Accessed 21 Feb 2018.
45. Vang R, Gown AM, Wu L-S-F, et al. Immunohistochemical expression of CDX2 in primary ovarian mucinous tumors and metastatic mucinous carcinomas involving the ovary: comparison with CK20 and correlation with coordinate expression of CK7. Mod Pathol. 2006;19(11):1421–8. https://doi.org/10.1038/modpathol.3800698.
46. Moh M, Krings G, Ates D, Aysal A, Kim GE, Rabban JT. SATB2 expression distinguishes ovarian metastases of colorectal and appendiceal origin from primary ovarian tumors of mucinous or endometrioid type. Am J Surg Pathol. 2016;40(3):419–32. https://doi.org/10.1097/PAS.0000000000000553.
47. Shah SP, Köbel M, Senz J, et al. Mutation of FOXL2 in granulosa-cell tumors of the ovary. N Engl J Med. 2009;360(26):2719–29. https://doi.org/10.1056/NEJMoa0902542.
48. Zhao C, Vinh TN, Mcmanus K, Dabbs D, Barner R, Vang R. Identification of the most sensitive and robust immunohistochemical markers in different categories of ovarian sex cord-stromal tumors. https://insights.ovid.com/pubmed?pmid=19033865. Accessed 1 Feb 2018.
49. Ramos P, Karnezis AN, Craig DW, et al. Small cell carcinoma of the ovary, hypercalcemic type, displays frequent inactivating germline and somatic mutations in SMARCA4. Nat Genet. 2014;46(5):427–9. https://doi.org/10.1038/ng.2928.
50. FIGO Committee on Gynecologic Oncology. Current FIGO staging for cancer of the vagina, fallopian tube, ovary, and gestational trophoblastic neoplasia. Int J Gynaecol Obstet. 2009;105(1):3–4. http://www.ncbi.nlm.nih.gov/pubmed/19322933. Accessed 16 Feb 2018.
51. Pecorelli S. Revised FIGO staging for carcinoma of the vulva, cervix, and endometrium. Int J Gynaecol Obstet. 2009;105(2):103–4. http://www.ncbi.nlm.nih.gov/pubmed/19367689. Accessed 16 Feb 2018.
52. Havrilesky LJ, Cragun JM, Calingaert B, et al. The prognostic significance of positive peritoneal cytology and adnexal/serosal metastasis in stage IIIA endometrial cancer. Gynecol Oncol. 2007;104(2):401–5. https://doi.org/10.1016/j.ygyno.2006.08.027.
53. Sonoda Y, Zerbe M, Smith A, Lin O, Barakat RR, Hoskins WJ. High incidence of positive peritoneal cytology in low-risk endometrial cancer treated by laparoscopically assisted vaginal hysterectomy. Gynecol Oncol. 2001;80(3):378–82. https://doi.org/10.1006/gyno.2000.6079.
54. Zullo F, Palomba S, Falbo A, et al. Laparoscopic surgery vs laparotomy for early stage endometrial cancer: long-term data of a randomized controlled trial. Am J Obstet Gynecol. 2009;200(3):296.e1–9. https://doi.org/10.1016/j.ajog.2008.10.056.
55. Prat J. Staging classification for cancer of the ovary, fallopian tube, and peritoneum. Int J Gynecol Obstet. 2014;124(1):1–5. https://doi.org/10.1016/j.ijgo.2013.10.001.
56. Heintz APM, Odicino F, Maisonneuve P, et al. Carcinoma of the ovary. Int J Gynecol Obstet. 2003;83(S1):135–66. https://doi.org/10.1016/S0020-7292(03)90118-4.
57. Fadare O, Mariappan MR, Hileeto D, Wang S, McAlpine JN, Rimm DL. Upstaging based solely on positive peritoneal washing does not affect outcome in endometrial cancer. Mod Pathol. 2005;18(5):673–80. https://doi.org/10.1038/modpathol.3800342.
58. Anastasiadis PG, Romanidis KN, Polichronidis A, Koutlaki NG, Tamiolakis D, Simopoulos K. The contribution of rapid intraoperative cytology to the improvement of ovarian cancer staging. Gynecol Oncol. 2002;86(3):244–9. http://www.ncbi.nlm.nih.gov/pubmed/12217743. Accessed 17 Feb 2018.
59. Fadare O, Mariappan MR, Wang S, Hileeto D, Mcalpine J, Rimm DL. The histologic subtype of ovarian tumors affects the detection rate by pelvic washings. Cancer. 2004;102(3):150–6. https://doi.org/10.1002/cncr.20239.
60. Bentrem D, Wilton A, Mazumdar M, Brennan M, Coit D. The value of peritoneal cytology as a preoperative predictor in patients with gastric carcinoma undergoing a curative resection. Ann Surg Oncol. 2005;12(5):347–53. https://doi.org/10.1245/ASO.2005.03.065.
61. Kanellos I, Demetriades H, Zintzaras E, Mandrali A, Mantzoros I, Betsis D. Incidence and prognostic value of positive peritoneal cytology in colorectal cancer. Dis Colon Rectum. 2003;46(4):535–9. https://doi.org/10.1007/s10350-004-6595-0.
62. Yamamoto S, Akasu T, Fujita S, Moriya Y. Long-term prognostic value of conventional peritoneal cytology after curative resection for colorectal carcinoma. Jpn J Clin Oncol. 2003;33(1):33–7. https://doi.org/10.1093/jjco/hyg007.
63. Castillo CF-D, Rattner DW, Warshaw AL. Further experience with laparoscopy and peritoneal cytology in the staging of pancreatic cancer. Br J Surg. 1995;82(8):1127–9. https://doi.org/10.1002/bjs.1800820839.
64. Makary MA, Warshaw AL, Centeno BA, Willett CG, Rattner DW, Castillo CF. Implications of peritoneal cytology for pancreatic cancer management. Arch Surg. 1998;133(4):361. https://doi.org/10.1001/archsurg.133.4.361.
65. Pretorius RG, Lee KR, Papillo J, Baker S, Belinson J. False-negative peritoneal cytology in metastatic ovarian carcinoma. Obstet Gynecol. 1986;68(5):619–23. http://www.ncbi.nlm.nih.gov/pubmed/3763072. Accessed 17 Feb 2018.
66. Cheng L, Wolf NG, Rose PG, Rodriguez M, Abdul-Karim FW. Peritoneal washing cytology of ovarian tumors of low malignant potential: correlation with surface ovarian involvement and peritoneal implants. Acta Cytol. 1998;42(5):1091–4. http://www.ncbi.nlm.nih.gov/pubmed/9755663. Accessed 17 Feb 2018.
67. Cibas ES. Peritoneal washings. In: Cibas ES, Ducatman BS, editors. Cytology: diagnostic principles and clinical correlates. 4th ed. Philadelphia, PA: Elsevier; 2014. p. 155–69.
68. Sneige N, Fernandez T, Copeland LJ, Katz RL. Müllerian inclusions in peritoneal washings. Potential source of error in cytologic diagnosis. Acta Cytol. 1986;30(3):271–6. http://www.ncbi.nlm.nih.gov/pubmed/3012920. Accessed 17 Feb 2018.
69. Slade AJ, Dieterich M, Sturgis CD. Eosinophilic metaplastic atypia in exfoliated cells of ovarian endometriosis: a potential cytodiagnostic pitfall in peritoneal fluids. Diagn Cytopathol. 2004;31(2):123–5. https://doi.org/10.1002/dc.20031.
70. Sidawy MK, Silverberg SG. Endosalpingiosis in female peritoneal washings: a diagnostic pitfall. Int J Gynecol Pathol. 1987;6(4):340–6. http://www.ncbi.nlm.nih.gov/pubmed/3692676. Accessed 17 Feb 2018.
71. Sams VR, Benjamin E, Ward RH. Ectopic pancreas. A cause of false-positive peritoneal cytology. Acta Cytol. 1990;34(5):641–4. http://www.ncbi.nlm.nih.gov/pubmed/2145720. Accessed 17 Feb 2018.
72. Jacques SM, Selvaggi SM. Multiple peritoneal cytologies collected during laparotomy for gynecologic malignancy. Diagn Cytopathol. 1991;7(5):482–6. https://doi.org/10.1002/dc.2840070509.
73. Soslow RA. Histologic subtypes of ovarian carcinoma. Int J Gynecol Pathol. 2008;27(2):161–74. https://doi.org/10.1097/PGP.0b013e31815ea812.

74. Zhang Y, Garcia-Buitrago MT, Koru-Sengul T, Schuman S, Ganjei-Azar P. An immunohistochemical panel to distinguish ovarian from uterine serous papillary carcinomas. Int J Gynecol Pathol. 2013;32(5):476–81. https://doi.org/10.1097/PGP.0b013e31826ddc4e.
75. Kounelis S, Kapranos N, Kouri E, Coppola D, Papadaki H, Jones MW. Immunohistochemical profile of endometrial adenocarcinoma: a study of 61 cases and review of the literature. Mod Pathol. 2000;13(4):379–88. https://doi.org/10.1038/modpathol.3880062.
76. Yemelyanova A, Vang R, Kshirsagar M, et al. Immunohistochemical staining patterns of p53 can serve as a surrogate marker for TP53 mutations in ovarian carcinoma: an immunohistochemical and nucleotide sequencing analysis. Mod Pathol. 2011;24(9):1248–53. https://doi.org/10.1038/modpathol.2011.85.