

# Current Challenges and Opportunities for Research on Borderline Ovarian Tumors

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This article summarizes key issues for future research on borderline ovarian tumors that emerged at a National Cancer Institute–sponsored Borderline Ovarian Tumor Workshop held in August 2003 in Bethesda, MD. Limitations in existing research and opportunities for future advances have been highlighted. The application of new molecular techniques in combination with improved study designs holds promise for elucidating the pathogenesis of these tumors and revealing the source of the extra-ovarian lesions (“implants”) with which they are frequently associated. Clarification of the etiology of borderline tumors and the pathogenesis of their associated implants

A substantial body of literature on the clinical and pathological features of borderline ovarian tumors (BOTs) has provided the basis for expert consensus on some key issues related to the diagnosis and management of these neoplasms (see the article by Silverberg et al elsewhere in this issue).<sup>1</sup> Nonetheless, disagreements and uncertainties about the etiology, pathogenesis, diagnosis, and management of BOTs persist. Some previously intractable questions can now be addressed using new laboratory techniques. Toward that end, this review has 2 main purposes: (1) to identify limitations of existing research on BOTs and suggest general strategies to remedy these deficiencies in future investigations, and (2) to identify a set of critical unanswered questions related to the biology and behavior of these tumors, emphasizing issues particularly relevant to clinical practice.

This discussion is restricted to serous BOTs (S-BOTs) and mucinous BOTs (M-BOTs), the specific histological tumor types that account for nearly all BOTs. The

is critical for improving pathological diagnosis, revising the classification system of ovarian neoplasms, and developing optimal, evidence-based clinical management algorithms. *HUM PATHOL* 35: 961-970. © 2004 Elsevier Inc. All rights reserved.

*Key words:* borderline, low malignant potential, ovary, neoplasm, epidemiology, pathology.

*Abbreviations:* BOT, borderline ovarian tumor; M-BOT, mucinous borderline ovarian tumor; S-BOT, serous borderline ovarian tumor; SEER, Surveillance, Epidemiology, and End Results.

views summarized in this review were developed during the National Cancer Institute–sponsored Borderline Ovarian Tumor Workshop held on August 27 and 28, 2003 in Bethesda, MD and in subsequent discussions among attendees.

## LIMITATIONS OF CURRENT RESEARCH ON BOTs

### General Observations

The clinical literature on BOTs has been dominated by reports of retrospective case series, leading to concerns regarding both the internal and external validity of reported data.<sup>2</sup> Few studies have been conducted in well-defined populations, and obtaining access to biological samples has presented substantial challenges. An overview of these considerations is summarized here (Table 1).

### Limitations of Existing Research on BOTs: Internal Validity

Most studies of BOTs have been retrospective, which has limited both the quality and quantity of data collected. In the comprehensive review by Seidman and Kurman,<sup>3</sup> which included 97 reported studies of 4129 patients with S-BOTs, only 6 prospective randomized studies of 373 patients were identified. Similarly, a literature search restricted to clinical trials based on the key words “ovary,” “mucinous,” and “borderline,” did not retrieve any citations. Retrospective studies often necessitate exclusion of cases when clinical data or diagnostic tissues are missing. The exclusion of a subset of cases can bias results when the cause for exclusion is associated with the outcome of interest (eg, recurrence or death). (Note that for the purpose of this discussion, “internal validity” refers to verification that the data from a specific investigation accurately represents the patients or samples studied, and “external validity” re-

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**TABLE 1.** Common Limitations in Studies of Borderline Ovarian Tumors**Internal validity**

- Retrospective designs with potential exclusion biases
- Pathology reviews unmasked
- Inadequate statistical analysis

**External validity**

- Referral biases
- Nonstandardized processing and reporting of pathology
- Unknown diagnostic reproducibility
- Lengthy case accrual may obscure changes in disease patterns

**Other concerns**

- Unreliable preoperative diagnosis complicates study designs
- Most cases diagnosed and initially treated in nonresearch settings
- Registry data limited; concerns about misclassification of cases
- No experimental models

flects the certainty that the results of a specific study accurately reflect the nature of the disease in the source population.)

The interpretations and descriptions reported in pathological studies have almost invariably reflected the opinions of a single expert (and his or her associates), rather than the more diverse views of a multimember panel representing the breadth of expertise in the field. This limitation is compounded by the difficulty of obtaining independent intradepartmental opinions retrospectively, given that controversial cases have frequently been reviewed and discussed among colleagues before sign-out.

Finally, most studies of BOTs have not used appropriate statistical methods, including calculation of person-time for follow-up data and multivariate analysis for identifying the independence of associations between pathological or clinical features and outcomes. Consequently, these analyses often do not provide useful estimates of absolute and relative risks of unfavorable outcomes, which represent a central factor in making informed treatment decisions.

#### Limitations of Existing Research on BOTs: External Validity

The literature on BOTs is based largely on studies performed in tertiary referral centers, which tend to treat more aggressive and/or unusual cases than those treated in community practice. Concerns about external validity are magnified when cases initially encountered in daily practice are combined with referral cases in the same analysis. Inclusion of the latter may enrich the study population with problematic cases without capturing a proportionate number of cases more representative of the disease in the community.

Although diagnostic criteria for BOTs have been described and illustrated,<sup>4,5</sup> many of these features have not been precisely defined, and thus it is unlikely that they have been uniformly applied by pathologists. It is unclear how well pathologists currently practicing agree on the diagnosis of BOTs. Previously, a histopathology review of a population-based series of 477 ovarian tumors diagnosed from 1980 to 1982 found that experts reclassified 15% of ovarian carcinomas re-

ported in the community as BOTs and 7% of BOTs as carcinomas.<sup>6</sup> Similarly, an audit of 64 cases of BOT referred to a UK institution with special expertise in gynecologic pathology between 1988 and 1997 found that 27 (42%) were misdiagnosed as ovarian carcinoma.<sup>7</sup> In another review of cases originally diagnosed as stage 1 or 2 ovarian carcinoma at a tertiary center (1980-2000), 29% of cases were reclassified as BOTs. Importantly, only 4.5% of women with tumors reclassified as BOTs died of disease as compared to 25.6% of patients with confirmed diagnoses of carcinoma.<sup>8</sup>

Minimal criteria for distinguishing subtle noninvasive implants from endosalpingiosis and florid noninvasive implants from invasive implants have neither been standardized nor rigorously validated.<sup>9,10</sup> Generally, in community practice pathologists favor more severe pathological diagnoses.<sup>6,7</sup> As a result, BOTs and BOTs associated with implants diagnosed in general practice may be associated with a better prognosis than those reported in the literature. Finally, given the relative rarity of BOTs, many reports are based on cases accumulated over long periods, which raise external validity concerns related to shifts over time in etiologic exposures, detection methods, diagnostic criteria, and treatments that could affect clinical presentations and behavior.

#### Limitations of Existing Research on BOTs: Other Considerations

An important limitation in studying BOTs is that a definitive diagnosis requires detailed histopathologic examination, which thus precludes preoperative diagnosis. Accordingly, it is logistically challenging, if not impossible, to prospectively obtain informed patient consent and collect data and specimens using a standardized protocol. Given that BOTs represent only a small fraction of adnexal masses, it would be expensive and inefficient to conduct a study of BOTs by recruiting cases from this large group of women with heterogeneous conditions. In addition, most BOTs are diagnosed in community institutions by generalists rather than in research centers by specialists. In one report, 78% of S-BOTs were initially managed by general obstetricians/gynecologists, whereas 10% were first treated by gynecologic oncologists.<sup>11</sup> Staging biopsies were done in 95% of operations performed by gynecologic oncologists, compared with only 65% of those performed by obstetricians/gynecologists, suggesting that some of the latter cases may have been understaged.

Expert review of histopathology notwithstanding, nonstandardized guidelines for processing tissues and reporting pathology may limit confidence in the adequacy of tissue sampling for microscopy, assessment of surface involvement, and evaluation of other factors. Destruction of unused portions of specimens and lack of access to high-quality photographs further limit research on the macroscopic features of these tumors. Incomplete cataloging of these images limits opportunities for understanding the growth and development

of BOTs and for developing educational resources that could be used by radiologists and pathologists.

Research on BOTs has been limited by the lack of relevant cell/tissue culture systems and animal models, which have proved useful in understanding the molecular pathogenesis of other tumors. The recent decision to discontinue reporting of BOTs in the U.S. Surveillance, Epidemiology, and End Results (SEER) program has eliminated a potentially valuable mechanism for identifying population-based cases in the United States. Hopefully, some individual registries will continue tracking these cases outside of the SEER program.

### Opportunities for New Approaches in the Study of BOTs

Incompletely standardized diagnostic criteria and use of different terminology represent barriers to conducting large, multicenter studies of BOTs. Particular points of concern include (1) the lack of minimal semiquantitative criteria for distinguishing cystadenomas from BOTs, (2) the absence of criteria for distinguishing BOTs from carcinomas without recognizable destructive invasion in the ovary (a diagnosis invoked by some pathologists based on malignant cytology), (3) the unconfirmed validity of reclassifying BOTs with substantial micropapillary growth (a minimum threshold of 5 mm has been suggested) as carcinomas, (4) variable terminology for distinguishing M-BOTs from M-BOTs with intraglandular carcinoma, and (5) imprecise criteria for distinguishing among endosalpingiosis, noninvasive implants, and invasive implants (see the sections on future directions below). Potentially, increased standardization of diagnostic criteria for BOTs could be established through organized microscopic reviews of shared cases among experts conducted either at scientific meetings or by mail. Recently, a website presenting a survey of practice patterns and images for independent classification was used to successfully assess the updated 2001 Bethesda System.<sup>12</sup> A similar site for BOTs might be useful. As a minimum, this approach could define interobserver reproducibility, its relationship to practice experience, and the degree to which inconsistencies reflect imprecise criteria as opposed to subjective judgments about microscopic appearances.

Pilot studies should be considered to determine the feasibility of studying samples of population-based cases identified through existing registries within the United States or abroad or by developing a new registry. If such studies prove practical, they might logically be combined with investigations of stage I carcinomas. Cooperation with patient advocacy groups, use of internet sites, and other mechanisms of targeted communication have shown promise for recruiting study subjects with rare diseases, although selection biases inherent in these approaches must be kept in mind. Studies performed in large health maintenance organizations with stable subscription bases may also provide opportunities for developing case-control studies and case-case

comparisons nested within well-defined retrospective cohorts.

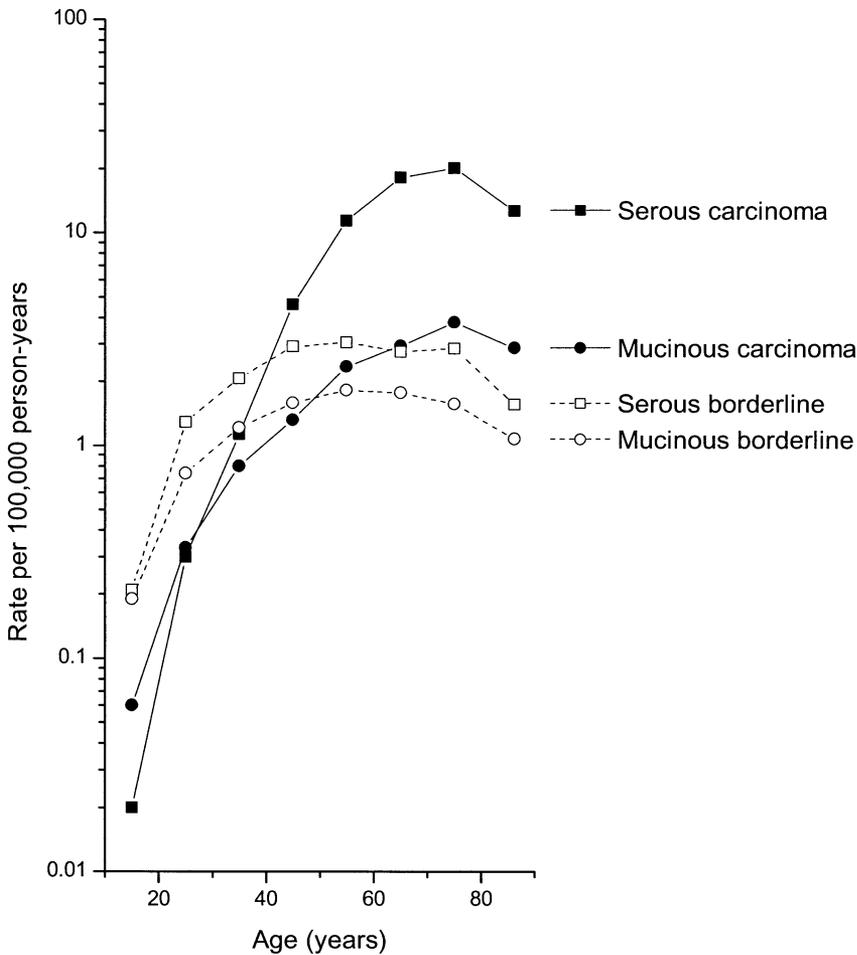
### OPPORTUNITIES FOR APPLYING NEW LABORATORY TECHNIQUES IN THE STUDY OF BOTs

Molecular techniques could potentially elucidate several key questions related to the pathogenesis of BOTs and their relationship to other ovarian tumors and lesions found in müllerian epithelium. Central questions include (1) whether a subset of BOTs progress to invasive carcinoma, and if so, how this subset may be identified; (2) whether extra-ovarian lesions represent spread from an ovarian primary, lesions that have developed de novo outside of the ovary (eg, peritoneum, fallopian tube), or a mixture of both; and (3) the pathobiological relationship of S-BOTs with micropapillary growth to other ovarian tumors. Preliminary answers are available for some of these questions, whereas others remain more controversial.

Molecular investigation of BOTs is challenging; primary tumors are generally rich in stroma and histologically heterogeneous, and implants are often small and paucicellular. High-resolution microdissection (eg, laser capture microdissection), followed by extraction and analysis of relevant macromolecules, could help overcome these limitations, particularly if high-fidelity amplification methods are developed and used. Assays such as immunohistochemistry or in situ hybridization that allow cellular and even subcellular localization of molecules of interest within intact tissue sections will undoubtedly continue to prove useful in studying BOTs. Other challenges for improving our understanding of the molecular pathogenesis of BOTs include the limited number of molecular alterations known to occur in these tumors,<sup>13-15</sup> the small number of alterations typically found per individual tumor, and the low growth fraction. Profiling strategies using expression arrays (cDNA- or oligonucleotide-based) or comparative genomic hybridization potentially could be used to search broadly for genetic abnormalities in these tumors, provided that valid amplification techniques can be used. Some groups have already used interphase cytogenetic techniques as a means of characterizing chromosomal aberrations without preparing traditional metaphase spreads.<sup>13,14</sup> Studies of "candidate markers" related to processes that would seem likely to predict aggressiveness, such as invasion, adhesion, and epithelial-stromal interactions, may also prove useful.<sup>15</sup>

### FUTURE RESEARCH DIRECTIONS: EPIDEMIOLOGY OF BOTs

Data from SEER demonstrate that BOTs are uncommon tumors with an incidence of  $\sim 2.5/10^5$  women-years; rates are lower in blacks than in whites, mirroring the relative rates for ovarian carcinoma in these groups.<sup>17</sup> Age-specific incidence rates for BOTs in-



**FIGURE 1.** Incidence of ovarian borderline and invasive serous and mucinous tumors in the United States. Note that rates of borderline tumors fail to rise after menopause, whereas rates for carcinoma continue to increase.

crease into the sixth decade of life and then stabilize, possibly declining among women in the ninth decade. Although rates for S-BOTs are slightly higher than those for M-BOTs, the overall age-specific patterns are similar. In contrast, rates of serous and mucinous carcinomas continue to increase for approximately 2 decades after menopause, with a much higher relative incidence for serous carcinomas; for unknown reasons, mucinous carcinomas are rare (Fig 1). Differences in age-specific rate patterns may imply differences in etiology. For example, the plateau in rates for BOTs around menopause may reflect an important effect of premenopausal exposures in the etiology of these neoplasms. In contrast, the steady increase in rates of carcinomas with increasing age would be more consistent with cumulative DNA damage resulting from total lifetime exposure to etiologic factors. In any case, the distinct age-specific rates for BOTs and carcinomas may represent one of the strongest clues that these tumors differ etiologically.

Incidence data from other countries and regional differences within the United States have received comparatively little attention, but could potentially provide useful etiologic clues (Table 2). One study found similar rates of ovarian carcinoma in Asian women born in the United States and those born in Asia, and for both

groups, lower rates than for whites in the United States.<sup>18</sup> These data suggest that a genetic or nongenetic factor prevalent among Asians is protective against ovarian carcinoma; analysis of similar data for BOTs would be useful.

Based on limited data, most risk factors for BOTs and invasive ovarian carcinomas generally seem similar,<sup>19,20</sup> with the notable exception that oral contraceptive use, which clearly reduces a woman's risk for developing carcinoma, may not affect the risk of developing BOT. Admittedly, these interpretations are limited by pathological misclassification of BOTs as ovarian carcinomas and metastases as M-BOTs. Risk factors associated with recurrence of noninvasive implants or with the development of invasive implants have not been determined. Identification of such factors could suggest preventive strategies for conservatively treated patients who may have undetected peritoneal disease and for developing adjuvant treatments for advanced disease. Alternative nonsurgical treatments are needed for these women, because conventional adjuvant chemotherapy does not seem to provide benefit after successful cytoreduction and may cause considerable morbidity.<sup>21-24</sup>

Studies are also needed to determine whether risk factors differ among women with BOTs stratified by

**TABLE 2.** Opportunities for Future Research on Borderline Ovarian Tumors**Epidemiology**

- Comparing risk factors for borderline tumors and carcinomas
- Identifying risk factors for invasive implants, recurrences, and death
- Identifying predisposing heritable factors

**Clinical management**

- Improved methods for preoperative and intraoperative diagnosis
- Determining risks associated with conservative treatment
- Role of laparoscopic surgery in removal of complex cysts
- Development of optimal standardized follow-up protocols
- Improved methods for distinguishing metastases from borderline tumors

**Pathology and molecular biology**

- Descriptive studies of the gross pathology of small borderline tumors
- Development of more precise reproducible criteria for distinguishing cystadenomas, borderline tumors, and carcinomas
- Interobserver reproducibility studies based on revised criteria
- Assessment of the accuracy of frozen section diagnosis and identifying means for improving performance
- Molecular studies to assess the relationship between serous borderline tumors, low-grade, serous carcinoma, and high-grade serous carcinoma
- Molecular studies to aid in the characterization of implants
- Comprehensive investigations to determine the incidence and behavior of serous borderline tumors with micropapillary features
- Studies to determine the biology of microinvasive borderline tumors and their clinical significance in long-term follow-up
- Improved and standardized criteria for separating endosalpingiosis, noninvasive implants, and invasive implants

pathological features, including cellular differentiation (serous vs mucinous), size, stage, laterality, surface involvement, and growth patterns (gross and microscopic). In particular, it is unclear whether implants of S-BOT reflect a “field effect” in which multiple synchronous and/or metachronous primary tumors develop from ovarian surface epithelium and/or other sites, or spread from a single primary ovarian tumor. Analysis of risk factors should include a pathology review when possible, especially to exclude misclassification of metastases to the ovary as M-BOT. Patients with M-BOTs are more frequently diagnosed with gastrointestinal cancers than women in the general population, but it is likely that some of these women presented with a metastasis to the ovary (possibly associated with pseudomyxoma peritonei) that was misdiagnosed as M-BOT. It is also possible that some fatalities ascribed to M-BOTs actually represent death due to metastases from undiagnosed nonovarian primaries (SEER data, 1992-2000).<sup>25</sup>

Germline mutations in *BRCA1* and *BRCA2* substantially increase the risk for ovarian carcinomas (and other tumor types), but it is unclear whether these mutations increase the risk of BOTs. Some studies have identified cases of BOT among carriers of germ line *BRCA* mutations,<sup>26,27</sup> but others have not.<sup>28</sup> Given that any increased risk of BOTs among carriers of *BRCA* mutations is likely to be relatively small compared with that for ovarian carcinomas, this question may be difficult to resolve. Of greater interest would be whether mutations in *BRCA* are related to the pathogenesis of

BOTs. Molecular studies to assess whether BOTs that occur among carriers have somatic inactivation of the wild-type *BRCA* allele in the tumor tissue might clarify this question. Identification of families in which multiple members have developed BOTs may also be valuable in exploring the development of these tumors.

**FUTURE DIRECTIONS: CLINICAL MANAGEMENT OF BOTs**

The development of better preoperative and intraoperative methods to distinguish benign ovarian cysts from BOTs and carcinomas would be extremely useful (Table 2). Notably, preoperative diagnosis would improve the selection of the surgical approach (intra-abdominal vs laparoscopic) and help plan the extent of surgery (cystectomy, oophorectomy, or a more extensive procedure, including staging), pending unexpected findings at exploration. Although studies of newer radiologic methods for detecting ovarian carcinomas have received considerable attention, the performance of these methods for identifying BOTs has not been fully explored, and future work would be useful.<sup>29</sup> Potentially, radiologic methods that could be used during surgery to complement intraoperative pathology consultations and frozen section diagnosis could be developed. Identification of serum markers specific for BOTs represents another possible area for future research.

Although women with BOTs who have completed childbearing often elect to undergo bilateral oophorectomy, young women may seek conservative treatment to preserve their fertility and maintain their premenopausal hormonal milieu. Clinical data suggest that conservative treatment (defined as retaining the uterus and at least part of an ovary) increases the risk of relapse compared with more definitive procedures. Based on a review of the literature, Morice et al<sup>30</sup> estimated that the risk of recurrence after conservative treatment is 0% to 20% and that after cystectomy the risk is 12% to 58%. However, these authors found that overall survival was not compromised by conservative treatment even when noninvasive implants were present and that many conservatively treated patients have successfully carried pregnancies to term.

Studies assessing the relative risk of recurrence and death among women treated conservatively have been limited by small size, limited duration of follow-up, and nonstandardized criteria for inclusion. Although recruitment of subjects for a randomized trial to assess different treatment options likely would be unsuccessful, a prospective study in which women selected their own treatments might still yield useful estimates of risk for unfavorable outcomes and data for quality-of-life measures. Such a study could also provide biological specimens that could be used to develop markers predictive of recurrence.

Several other issues related to the appropriateness of conservative management require additional research, including (1) the need for reoperation to per-

form staging when a diagnosis of BOT is made postoperatively, (2) the requirement to perform a bilateral oophorectomy after a successful pregnancy, and (3) the safety of fertility treatments among women treated conservatively. A recent retrospective analysis of clinical outcomes found that results were similar for women who were fully staged and those who underwent more limited surgery, but the authors noted that additional studies are needed.<sup>31</sup> Published data related to the safety of ovulation-inducing fertility treatments are also sparse.<sup>30</sup>

The estimated risks and benefits of laparoscopic surgery as compared with intra-abdominal surgery for women with radiologically identified complex cysts should be defined in controlled studies. It is anticipated that laparoscopic surgery would reduce the risks of adhesions and other complications, but increase the risk of cyst rupture and spillage. Based on retrospective review of published data, some authors have suggested that adhesions may represent an important cause of morbidity among some women with BOTs,<sup>32</sup> suggesting that procedures to limit this risk would be desirable. However, it is unclear whether cyst rupture would increase the risk for recurrence or death, and if so, whether specific surgical or medical interventions could be taken after spillage of cyst contents to minimize or eliminate these risks. In one retrospective analysis of 1545 women diagnosed with stage 1 ovarian carcinoma (based largely on hysterectomy, oophorectomy, and infracolic omentectomy without standardized staging), rupture was an important factor in predicting disease-free survival.<sup>33</sup> Further innovations to optimize laparoscopic techniques for removing ovarian cysts would benefit many patients.

Improved histopathologic criteria and immunohistochemical assays for distinguishing mucinous adenocarcinoma metastatic to the ovary from primary M-BOT have been proposed, but this differential diagnosis remains problematic.<sup>34-37</sup> Investigations suggest that the presence of multiple lesions within an ovary, bilateral ovarian involvement, involvement of normal-sized ovaries, and the presence of lesions on the surface or hilum strongly favor a metastasis. However, a scholarly review prepared for the gynecologic and surgical literature might be useful, because these data are probably more widely appreciated among pathologists. Reviews for nonpathologists should emphasize that a metastasis to the ovary may mimic either a BOT (especially an M-BOT) or a primary ovarian carcinoma; the concept that a metastasis might be confused with a lesion less aggressive than carcinoma (ie, BOT) may seem counterintuitive to nonmorphologists.

Guidelines for which preoperative or intraoperative procedures should be considered to differentiate metastatic tumors from primary ovarian neoplasms, including BOTs, would be useful. The development of rapid immunostaining methods that could be performed on frozen sections to aid in this differential diagnosis might prove helpful. Guidelines regarding follow-up tests, the frequency of follow-up examinations, and the total duration of posttreatment surveil-

lance after a diagnosis of BOTs should be developed. The success of salvage therapy for recurrences after conservative treatment should be determined using appropriate statistical methods to estimate risk of morbidity and mortality, so that patients and caretakers can make informed decisions. Studies to determine the impact of conservative treatment on quality of life might assist patients in choosing among management options.

#### **FUTURE DIRECTIONS FOR RESEARCH ON THE PATHOLOGY AND MOLECULAR BIOLOGY OF BOTs**

The macroscopic appearance of BOTs has been described and illustrated,<sup>4,5</sup> but studies describing the gross appearance of partially involved ovaries and small BOTs may further our understanding of the pathogenesis of these neoplasms (Table 2). Issues for further analysis would include the relationship between BOTs and the surrounding uninvolved ovarian parenchyma, early patterns of loculation, and patterns of cystic distention into the pelvis. Computer analysis of images of gross photographs might provide data useful in developing improved diagnostic criteria for making the diagnosis of BOT on gross pathological examination or radiologic studies. The mechanisms of fluid accumulation in BOTs and the biophysical effects of tension secondary to cyst distention have received little attention, although mechanical factors are known to effect gene expression in some tissues, and thus these processes could affect both the development and natural history of these tumors. In particular, the mechanical effects of cyst distention might alter the expression of adhesion molecules or reduce cell viability and growth. Intratumoral comparisons of cystic and noncystic areas might provide data about these issues. Analysis of cyst fluid collected by *ex vivo* puncture could aid in the discovery of secreted molecules that would be useful for diagnosis, staging, or detecting recurrences.

Multiple studies to identify microscopic distinctions between BOTs and ovarian carcinomas have been performed; however, criteria for distinguishing BOTs from cystadenomas have received less attention. The development of minimal criteria for BOTs based on data related to the risk of extra-ovarian disease would be clinically valuable, especially if more restrictive criteria permitted safe conservative management of women with cystadenomas containing minute proliferative foci that might be classified as BOTs by some pathologists. Changing minimal criteria for the diagnosis of BOT would require reassessment of clinical outcomes among women with more strictly defined tumors maintained within the BOT category. As with the distinction between BOT and cystadenoma, more morphological studies are needed to define the boundaries, distinctions, and differential diagnosis between categories, rather than publications that illustrate classic cases and document their clinical outcomes.

The definitive diagnosis of BOT is generally made

postoperatively; intraoperative frozen section diagnoses of BOT are provisional, pending further sectioning and microscopic examination. Even among experienced pathologists, frozen-section diagnoses of BOTs are often not confirmed on permanent sections; 2 studies found that frozen section diagnoses of BOT agreed with final diagnoses in only ~60% of cases.<sup>11,38</sup> A third study found that the frozen-section diagnosis of BOT is less accurate than that of other ovarian tumors; underdiagnosis was especially problematic for M-BOTs and BOTs weighing >1360 g.<sup>39</sup> Misdiagnosis of frozen sections may have an adverse impact on patient care, leading to overtreatment in some cases and failure to perform staging in others. Additional studies are needed to determine whether sampling of tissue for frozen section accounts for essentially all of these diagnostic errors, and, if not, other causes, such as diagnostic pitfalls, should be explored and illustrated.

Data indicate that women with BOTs tend to be older than those with cystadenomas but younger than those with carcinomas, which historically prompted speculations that the pathogenesis of carcinomas was related to progression of preexisting benign tumors. Currently, the general consensus among pathologists is that most serous carcinomas (ie, typical high-grade invasive carcinomas) do not arise from either cystadenomas or S-BOTs, a conclusion that is supported by some molecular studies.

One study reported that p53 alterations are not found in isolated S-BOTs or cystadenomas, but may be found in benign cysts associated with carcinomas.<sup>40</sup> However, this association could represent progression of cystadenomas to carcinomas, ingrowth and replacement of benign cyst epithelium by cancer, or distention and attenuation of carcinomatous cysts that results in a morphologically bland appearance mimicking cystadenoma. Other studies have found that p53 abnormalities are common in cancers but relatively rare in S-BOTs, whereas point mutations in *K-ras*, *B-raf*, and microsatellite instability are more characteristic of S-BOTs. Data also suggest that loss of heterozygosity is rare in S-BOTs compared with carcinomas.<sup>15,41-45</sup>

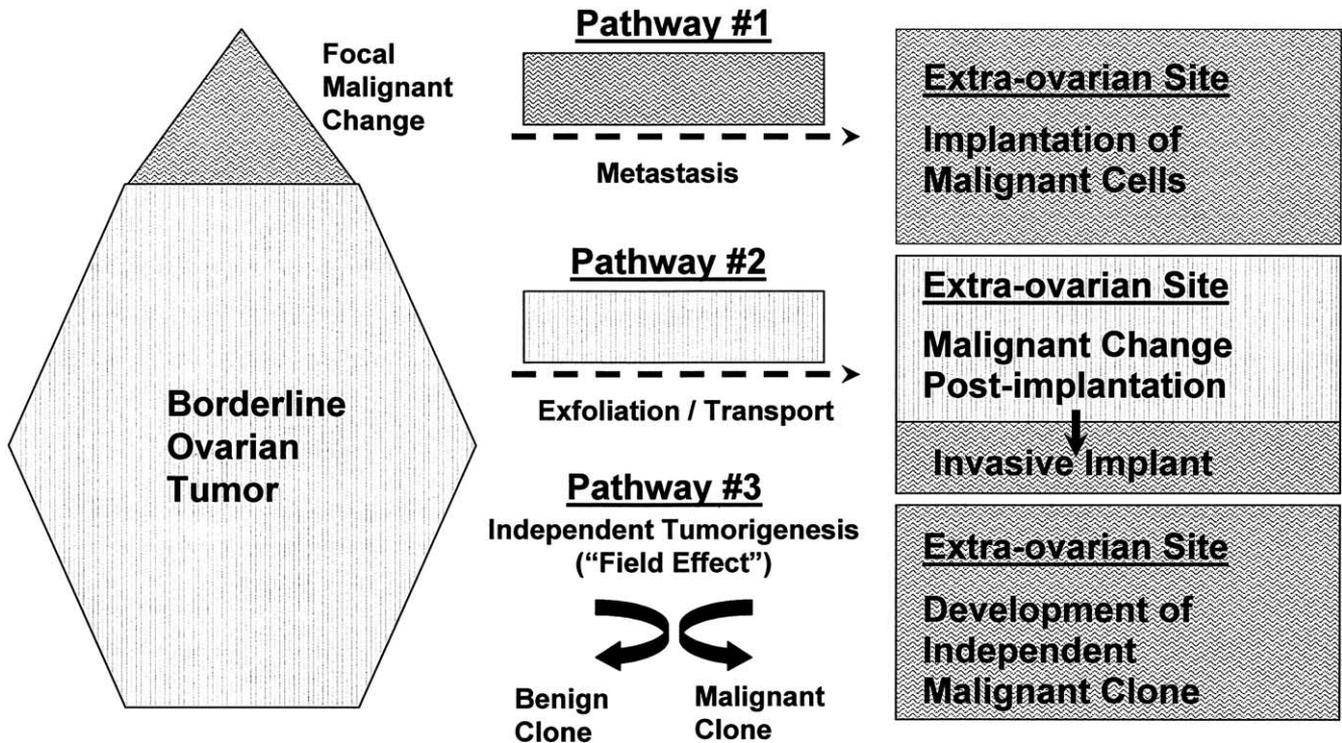
In contrast, detection of shared molecular alterations in S-BOTs and some serous carcinomas<sup>13,45-47</sup> might argue for a model in which at least a subset of S-BOTs progress to invasion. In particular, mutations in *K-ras* and *B-raf*,<sup>45</sup> trisomy 12,<sup>46</sup> and other cytogenetic abnormalities that have been identified in S-BOTs have also been found in low-grade or early-stage serous carcinomas, but not in high-grade tumors.

In contrast to serous ovarian carcinomas, there is general consensus that many mucinous ovarian carcinomas develop from a preexisting M-BOT. Foci suggesting in situ malignant change (“intraglandular carcinoma”) are often identified in M-BOTs associated with invasion, and the transition may be identified microscopically in some cases. Cytogenetic studies have demonstrated several aberrations common to M-BOT and invasive mucinous carcinomas, and these alterations may be found in both noninvasive and invasive components of some tumors.<sup>14,48</sup> However, molecular

data for M-BOTs are limited, and identifying molecular markers of invasion might have utility in selecting patients for staging. The pathogenesis of mucinous tumors that appear to arise from preexisting lesions, such as endometriosis and benign teratomas, is also of interest, but these cases have been studied mainly with light microscopy.

The description of S-BOTs with significant micropapillary growth and the view espoused by at least one group that these tumors should be reclassified as “micropapillary serous carcinoma” has sparked controversy.<sup>49-53</sup> As an initial step, semiquantitative, reproducible criteria for diagnosing these tumors should be developed. Issues related to the clinical importance of the micropapillary pattern would be most effectively resolved in a large interinstitutional or population-based study using a pathology review carefully masked to clinical outcomes and employing an appropriate statistical analysis. Specifically, the independent prognostic importance of the micropapillary pattern with regard to the risk of implants (noninvasive and invasive), recurrence, and death should be compared with that of other S-BOTs in a multivariate analysis controlling for other potential prognostic features, such as surface involvement and bilaterality. Many pathologists have not separated S-BOTs with a micropapillary pattern from other S-BOTs; thus these tumors are almost certainly underreported. Accordingly, the true incidence and behavior of S-BOTs with micropapillary growth in the population could differ from that in reported series.

The biologic and clinical significance of “microinvasion” and “involvement” of lymph nodes by BOT remain unclear. Seidman et al,<sup>5</sup> based on a literature review, found that survival was 100% among 94 patients with BOTs reported as demonstrating microinvasion (mean follow-up 7.4 years) and 98% among 43 women whose tumors reportedly involved lymph nodes (mean follow-up 6.5 years). Although these data are reassuring, long-term follow-up of such cases remains of interest, given that BOTs have been reported to recur more than a decade after initial diagnosis. Morphologically, microinvasion may appear as isolated cells with relatively abundant eosinophilic cytoplasm embedded in stroma or as small nests of cells, sometimes surrounded by clefts. It is uncertain whether these 2 patterns represent the same or different biological processes and whether the cells are actually producing lytic enzymes typical of invasion. Based on rare reported cases, microinvasion may be found more often in BOTs diagnosed during pregnancy, but limited follow-up has not identified more aggressive behavior of these tumors.<sup>54</sup> Similarly, the presence of BOT cells in lymph nodes could occur through primary spread from an ovarian primary tumor, secondarily from implants that have shed cells into peritoneal fluid that were later absorbed by lymphatics draining the peritoneal cavity, or by neoplastic change in benign intranodal glandular inclusions. It is also unknown whether these cells express genes that have been associated with adherence to vessels and invasion.



**FIGURE 2.** Selected pathways to explain the development of invasive implants in women with BOTs. Pathway 1: Borderline tumor undergoes malignant change in the ovary and spreads via a metastatic process. Pathway 2: Benign tumor cells arrive at an extra-ovarian site via a passive process such as mechanical detachment or transport via lymphatics; malignant change occurs after the benign cells have implanted. Pathway 3: Implants develop concurrently or metachronously from nonovarian epithelium and thus are not clonally related to the BOT.

More research on BOTs associated with invasive implants is needed to define pathological criteria, the population-based frequency, and the risks of recurrence and death. In a review by Seidman and Kurman,<sup>3</sup> invasive implants were associated with fatality in 35 of 104 women (34%). As noted for micropapillary S-BOTs, studies of invasive implants based on population-based samples should be performed. In addition, criteria for distinguishing noninvasive implants from endosalpingiosis and other benign proliferative lesions should be refined, standardized, and broadly disseminated to ensure that treatment-related morbidity secondary to overdiagnosis of implants and spurious upstaging of cases can be avoided, if in fact this is occurring. Detailed analyses of findings in fatal cases may also provide additional insights into disease progression and natural history. Pooling data from multiple centers may facilitate such studies.

**OPTIMIZING TERMINOLOGY FOR BOTs: WHAT'S IN A NAME?**

Ideal terminology should provide immediate insights into the biology and clinical behavior of the designated disease entity, thereby providing patients and clinicians with an understanding of management options and prognosis. The terms "borderline malignant" and "low malignant potential" both indicate

some possibility that a tumor will demonstrate malignant behavior, and the term "implant" suggests that the extra-ovarian lesion in question is derived from exfoliation of cells from an ovarian primary. The appropriateness of these terms depends on the pathogenesis of BOTs and their associated implants and the implications that these biological processes have for producing fatalities (Fig 2).

Based on data from the SEER, it is possible to argue that the relative survival of women with S-BOTs localized to the ovary approximates that of women without cancer in the general population.<sup>25,55</sup> This fact alone does not prove that S-BOTs are benign, because many overtly malignant tumors localized to the organ of origin are reliably cured by excision. Data also clearly demonstrate that survival in women with S-BOTs associated with implants is lower than that in women without cancer in the general population. However, this observation does not provide proof that S-BOTs are malignant. Two critical issues relevant to assessing these data are whether implants are derived from primary ovarian BOTs or develop de novo at extra-ovarian sites (eg, peritoneum, fallopian tube), and whether the cells that compose implants are biologically malignant and, if so, whether they have undergone transformation before or after reaching the extra-ovarian site. Implicitly, these issues are also related to whether cells reach

extra-ovarian sites by a process that could be characterized as “metastasis” or through another mechanism.

Data comparing the molecular signatures of S-BOTs and their associated implants are sparse and conflicting; some results suggest that these lesions are derived from the same clone, whereas others suggest clonal independence. Understanding the clonal relationships between BOTs and their associated implants would have important implications for refining diagnostic terminology.<sup>56-59</sup> If the cells of S-BOTs and their implants (especially invasive ones) are derived from separate clones, and if we accept that survival for stage I S-BOTs is essentially 100%, then the development of a new benign term for these tumors would be supported. However, if S-BOTs and their implants are clonally related, then optimal terminology should reflect the mechanism(s) that lead to the formation of the implants (Fig 2).

The fact that implants may be derived from ovarian primary tumors proves neither that the cells were malignant when they were deposited, nor that they reached the extra-ovarian site through a metastatic process. The biology of endometriotic lesions may provide a useful analogy; according to one viewpoint, endometriosis exhibits properties of a benign neoplasm, which has the exceptional capacity to spread throughout the body via a nonmetastatic mechanism and then undergo malignant change at sites distant from the uterus. Similarly, if it were demonstrated that implants were derived from the spread of ovarian tumor cells, this would not necessarily justify classifying the ovarian tumors as malignant, even in cases in which the implants appeared and behaved like carcinomas. It is possible that exfoliation or intravascular transport of benign ovarian tumor cells, or peritoneal metaplasia, could result in the formation of implants, whereas malignant transformation could be a distinct process that occurs at the extra-ovarian site. This issue might be addressed by comparing the molecular signatures of S-BOTs and of noninvasive and invasive implants removed from the same patient concurrently or sequentially. Finally, it is possible that several mechanisms account for the development of implants, perhaps justifying the maintenance of terminology that reflects biological and clinical ambiguity.

## CONCLUSION

The behavior of BOTs has been enigmatic, in part because it has been difficult to reconcile the histopathology of the tumors with the range of observed clinical outcomes. Expanded understanding of the etiology and pathogenesis of these tumors could have important implications for pathologic diagnostic criteria and terminology, clinical management and prognosis.

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