Modern Pathology (2010) 23, 1486–1491

Clinical outcome in diagnostically ambiguous foci of 'gland crowding' in the endometrium

Eric C Huang, George L Mutter, Christopher P Crum and Marisa R Nucci

Division of Women's and Perinatal Pathology, Department of Pathology, Brigham and Women's Hospital, Harvard Medical School, Boston, MA, USA

Premalignant endometrial lesions (endometrial intraepithelial neoplasia (EIN)) are clonal neoplasms that arise focally and can be diagnosed using specific criteria: (1) area of glands exceeds that of stroma (glands/stroma >1), (2) nuclear and/or cytoplasmic features of epithelial cells differ between architecturally abnormal glands and normal background glands, and (3) maximum linear dimension exceeds 1 mm. However, localized groups of crowded endometrial glands may be encountered that do not fulfill all of the criteria for EIN, are interpreted as ambiguous, and are reported as 'focal gland crowding'. We conducted a retrospective study of gland crowding using a free-text index search for this term in our pathology files. The age of the patients, number of subsequent specimens, the duration, and the outcome of the follow-ups were recorded. Of the 71 579 consecutive gynecological pathology reports, 206 (0.3%) 'gland crowding' cases were identified, in which 69% (143/206) had follow-up sampling. Of these, 33 (23%) had an outcome diagnosis of EIN (27 cases; 19%) or carcinoma (6 cases; 4%). Included were 18 cases (55%) diagnosed within the first year and presumed concurrent, and an additional 15 (45%) discovered after 1 year and interpreted as a later phase of disease or new events. The term 'crowded glands' is a highly significant finding that carries a substantial risk of an outcome of EIN and occasionally malignancy. It underscores the importance of follow-up when some but not all of the criteria for EIN are encountered in the appropriate clinical setting.
Modern Pathology (2010) **23,** 1486–1491; doi:10.1038/modpathol.2010.140; published online 30 July 2010

Keywords: endometrial hyperplasia; endometrial intraepithelial neoplasia; gland crowding

Endometrial carcinoma is the most common malignant tumor in the female genital tract with an age-adjusted incident rate of 23.5 per 100 000 women per year in the United States.[1] Patients often present with abnormal uterine bleeding that initiates endometrial sampling.[2–4] Clinical management of these patients has been based on the severity of an endometrial hyperplasia and reflects risk of concurrent or future endometrial carcinoma.[5–7] The World Health Organization (WHO) divides endometrial hyperplasia by architecture (simple or complex) and cytological atypia (with or without).[8] However, the reproducibility of this system among pathologists has been rather poor.[9,10] Endometrial intraepithelial neoplasia (EIN) has been introduced as an alternative term and has been adapted as an alternative schema to the WHO classification.[11,12] The diagnostic criteria for EIN include: (1) area of glands exceeds that of stroma (glands/stroma >1), (2) nuclear and/or cytoplasmic features of epithelial cells differ between architecturally abnormal glands and normal background glands, and (3) maximum linear dimension exceeds 1 mm. All of these features must be present to make the diagnosis. A diagnosis of EIN carries a substantial risk of concurrent or subsequent endometrial adenocarcinoma (25–35%).[13,14]

Although the criteria for EIN are well defined, we nevertheless occasionally encounter localized groups of cytologically altered crowded endometrial glands that may not satisfy all three criteria. These subdiagnostic samples are often interpreted as ambiguous and designated by the pathologist as 'gland crowding'. The differential diagnosis may include artifacts or mimics, and tangentially or poorly sampled neoplastic lesions. It has been our practice to flag these cases in the pathology report by describing them as 'gland crowding', and request repeat sampling to resolve this diagnostic dilemma. In this study, we report the clinical outcome and

Correspondence: Dr MR Nucci, MD, Division of Women's and Perinatal Pathology, Department of Pathology, Brigham and Women's Hospital, 75 Francis Street, Boston, MA, 02115, USA.
E-mail: mnucci@partners.org
Received 14 April 2010; revised and accepted 28 June 2010; published online 30 July 2010

significance of the term 'gland crowding' when applied in the context of the EIN criteria.

Materials and methods

EIN was first introduced as a diagnostic entity in January 2001 in the Division of Women's and Perinatal Pathology at the Brigham and Women's Hospital (Boston, MA, USA), a 700+ bed tertiary academic teaching hospital. To determine the clinical significance and outcome of 'gland crowding', we conducted a retrospective review of all gynecological surgical pathology cases from January 2001 to August 2009 following the introduction of EIN terminology at our institution. All reports were retrieved from the PowerPath anatomic pathology information system (IMPAC Medical System, CA, USA) using a combination of a free-text indexed search for the flagged term 'gland crowding' through the final anatomic diagnoses. The analysis included the age of the patients, number of subsequent specimens, the duration, and the outcome of the follow-ups. In addition, 26 cases (12 with benign and 14 with neoplastic follow-up samplings) were randomly selected and re-reviewed by three senior pathologists (CPC, GLM, and MRN) who were blinded to the outcome. Each pathologist was asked to predict, based on the histological features present in the area of gland crowding, whether the outcome was benign or neoplastic to potentially identify histological features that could identify those cases of 'gland crowding' in which there was a neoplastic outcome.

Results

A total of 71 579 gynecological specimens were surveyed, of which 206 (0.3%) had the term 'gland crowding' within the primary diagnostic field or associated note. Among the cases with crowded glands (Figures 1a and 1b, 2a and 2b, and 3a and 3b), 143 (69%) had follow-up pathology reports. Patient age ranged from 18 to 80 years, and the number of subsequent biopsies ranged from 1 to 16 (mean = 1; average = 1.8). In all, 73 (51%) and 130 (91%) patients received follow-up procedures within 6

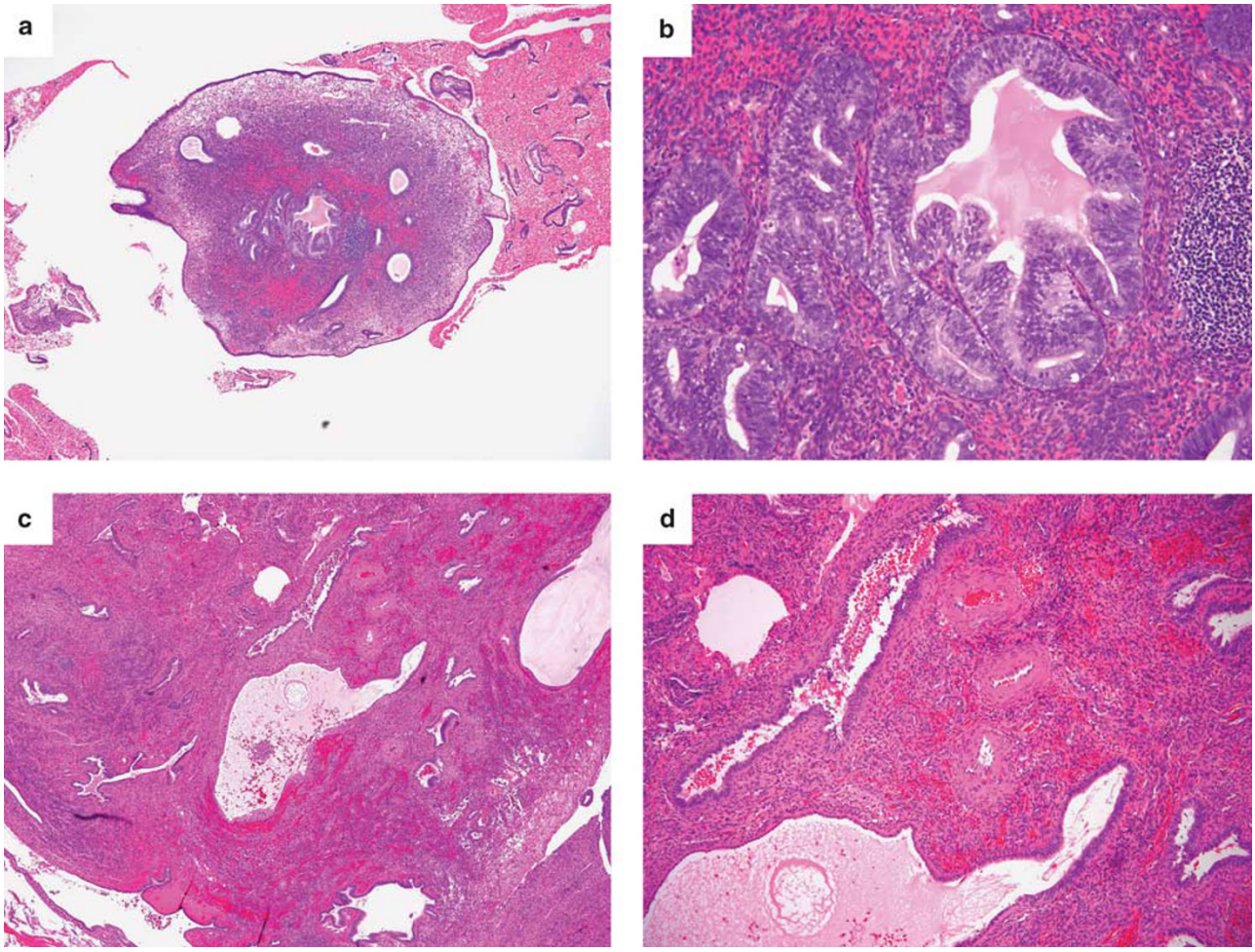

Figure 1 (**a**, **b**) Focal gland crowding (<1 mm) with cytological demarcation. (**c**, **d**) Follow-up sampling revealed an endometrial polyp with cystic glands, fibrous stroma, and thickened vessels.

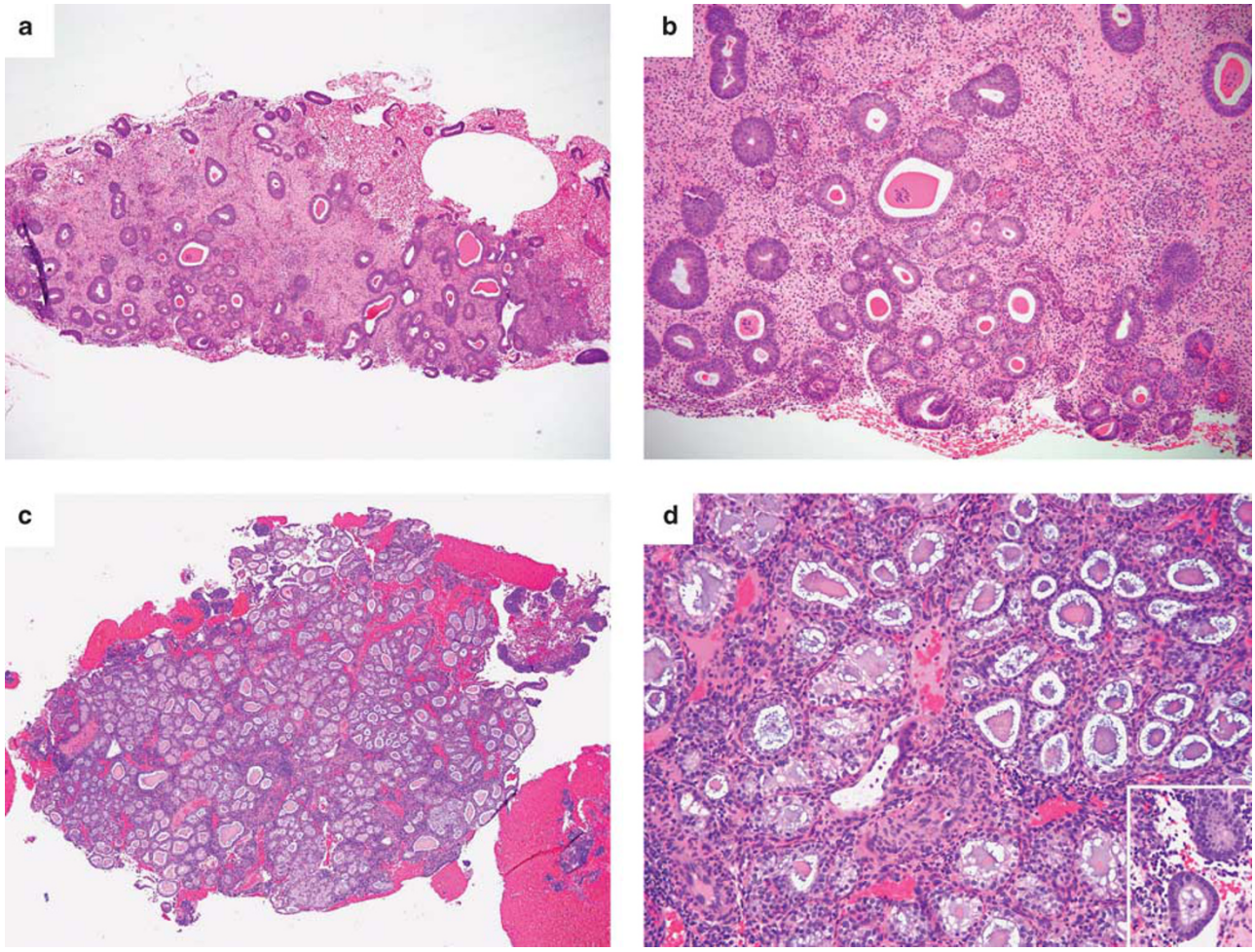

Figure 2 (**a**, **b**) Endometrial polyp with focal gland crowding. (**c**, **d**) Follow-up sampling showed endometrial intraepithelial neoplasia with crowded glands and cytological demarcation. Inset: background glands with normal cytology for comparison.

months and 1 year, respectively, after the initial report of 'gland crowding' (Figure 4). Nearly 77% of patients (110 cases) had a benign follow-up sampling (ie, proliferative endometrium, secretory endometrium, endometrial polyp, etc; Figure 1c and d) and 23% (33 cases) had subsequent diagnosis of neoplasia (Figure 5).

Among the flagged cases, 27 (19%) cases had a subsequent diagnosis of EIN with a duration of follow-up sampling ranging from 1 month to 7 years (median = 1 year; average = 1.5 years; Figure 2c and d). Carcinoma was identified in 6 (4%) cases (Figure 3c and d), ranging from 1 month to 5 years (median = 0.5 year; average = 1.7 years) after initial 'gland crowding'. In all, 14 (42%) EINs and 4 (12%) carcinomas were diagnosed within the first year and were presumed to be concurrent with the initial lesion. In addition, 13 (39%) additional EINs and 2 (6%) carcinomas occurred after 1 year and were interpreted as disease progression or new events. A total of 16 cases of gland crowding were initially identified within an endometrial polyp and of these, 11 cases had a benign follow-up, 4 had EIN, and 1 had carcinoma.

To decipher whether 'gland crowding' had morphological features that may suggest neoplastic progression, cases with benign, EIN, and carcinoma outcomes were randomly selected and re-reviewed by three senior pathologists. Possible worrisome features noted by pathologists include squamous morules, mucinous differentiation, nuclear stratification, papillary architecture, and intraglandular epithelial budding/tufting. However, none of the pathologists accurately predicted the outcomes (data not shown). Cases of 'gland crowding' showing any of these features can be followed by either benign or neoplastic outcomes.

Discussion

EIN is defined as a clonal proliferation of premalignant endometrial glands that are prone to malignant transformation to endometrial endometrioid adenocarcinoma.[15] Diagnostic criteria for EIN have been developed by histopathological correlation with clinical outcomes, molecular correlates (monoclonal growth, lineage continuity with carcinoma), and

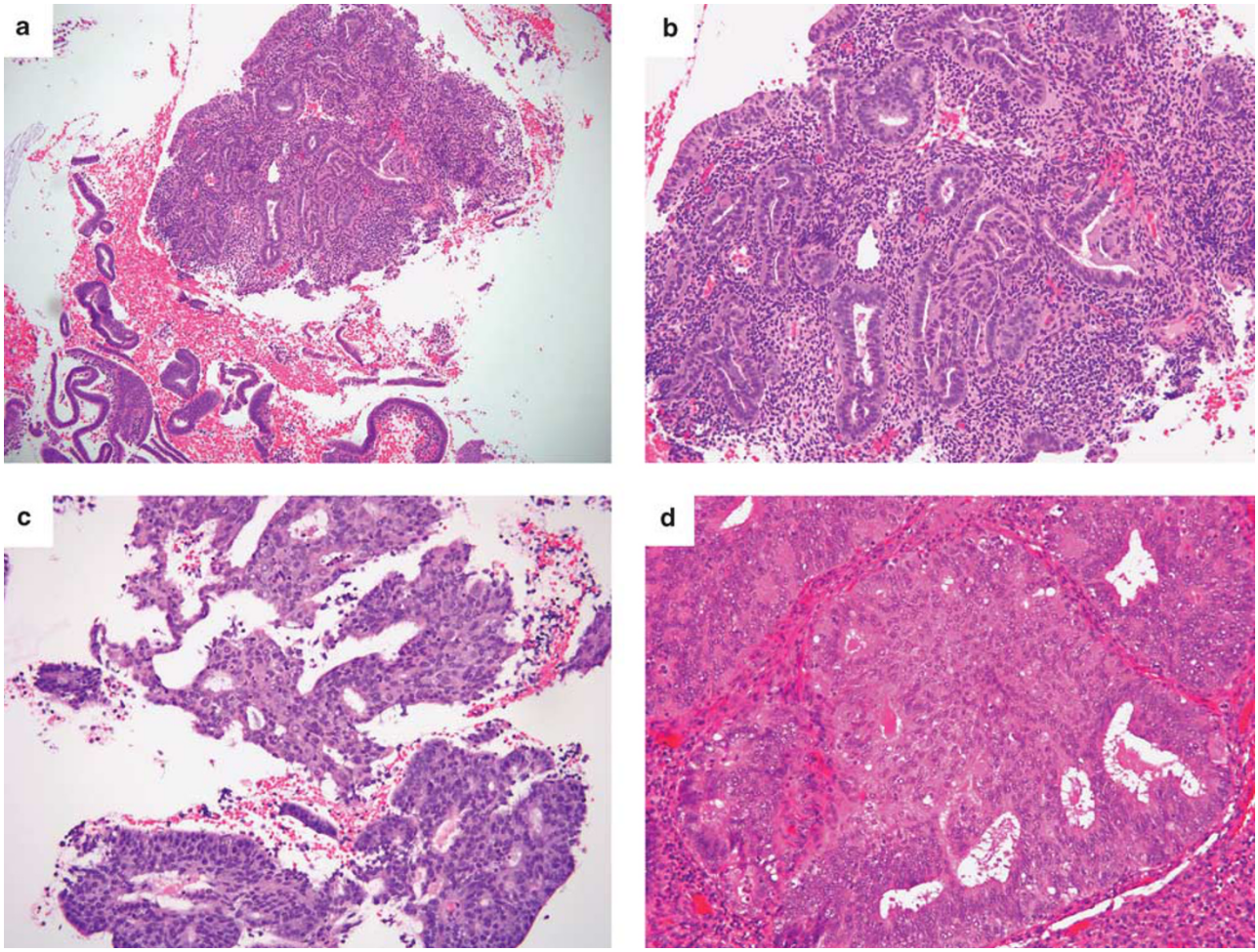

Figure 3 (**a**, **b**) Crowded glands distinct from background endometrium. (**c**) Follow-up sampling showed grade 1 endometrioid endometrial adenocarcinoma as indicated by the cribriform growth pattern. (**d**) Hysterectomy proved to be grade 2 endometrioid endometrial adenocarcinoma.

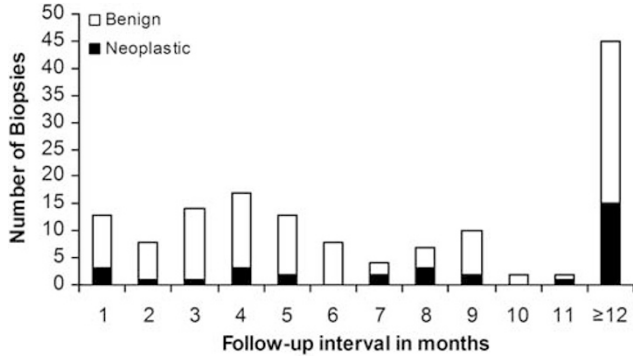

Figure 4 Clinical outcomes of follow-up biopsies by follow-up interval after an initial diagnosis of 'gland crowding' (143 total cases).

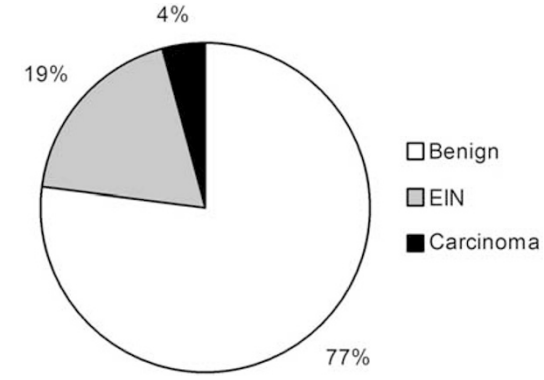

Figure 5 Follow-up outcome after initial 'gland crowding' (143 total cases).

objective computerized histomorphometry. As EINs emerge as clonal outgrowths, they can be readily recognized in routine practice by their architectural crowding and cytologically altered glands distinct from the background endometrium. This diagnostic strategy contrasts with the WHO 1994 classification system, which classifies these neoplastic precursors as endometrial hyperplasias and subdivides them by architectural complexity and cytological atypia.[16] To some, the hyperplasia schema, although an important improvement over the earlier and archaic terms for early endometrial neoplasia, has not been easily

reproduced across multiple observers or optimally stratified patients' cancer risk.[17]

Although the EIN classification system has offered some advantages over the hyperplasia system, most notably a clearer separation of precursor from background benign endometrial hyperplasia, lesions are still encountered that do not fulfill the criteria for EIN. These ambiguous cases do not represent a diagnostic class of presumed pre-EIN lesions, but rather diagnostically challenging cases that exceed the threshold of a diagnostically 'normal' biopsy. These are analogous to atypical squamous cells in cervical cytology, which in themselves are not a diagnostic entity but confer risk of neoplasia and require follow-up. These diagnostically ambiguous cases have been reported by us as having 'gland crowding' and follow-up sampling was typically recommended. To better understand the effect of this terminology on patient outcome and care, we sought to do a retrospective study on all cases that were identified as having 'gland crowding'.

In our practice, the use of the term 'gland crowding' was not common, occurring in just 0.3% of reports from 2001 to 2009. Among these 206 cases, 143 or almost 70% had follow-up sampling after an index report of 'gland crowding', in which 91% (130 of 143) occurred within the first year. In all, 23% (33 of 143) of the 'gland crowding' cases had a subsequent neoplastic process. Of these, 14 and 4 cases were diagnosed as EIN and carcinoma, respectively, within the first year. Two of the EIN cases were subsequently diagnosed as carcinoma in the hysterectomy specimens. These cases, based upon temporal proximity, are presumed to be concurrent lesions from the initial biopsy. In addition, 15 additional neoplastic cases (13 EINs and 2 carcinomas) were discovered more than a year from initial sampling. Two patients with carcinoma were diagnosed 3.5 and 5.5 years after the initial sampling. The first patient (46 years old) underwent a 7-month follow-up biopsy that proved to be proliferative endometrium and 3.5 years later developed grade 1 endometrial adenocarcinoma. The second patient (59 years old) had a report with recommendation for repeat sampling in 3 to 6 months; however, she returned 3 years later that showed benign endometrium. Two and a half years later, she developed grade 2 endometrial adenocarcinoma. These two cases suggest that neoplasms diagnosed after the first year may sometimes be newly developed lesions, unrelated to the initial crowded focus in the index sample.

It is imperative to emphasize that although 'gland crowding' is not a diagnostic entity, a descriptive reporting of gland crowding in diagnostically difficult cases is clearly warranted, as 23% of the patients in our series had a diagnosis of neoplasia in follow-up samples. The presence of crowded glands within a polyp (*vs* non-polypoid endometrium) did not result in a lower incidence of neoplasia on follow-up sampling as 31% of cases of gland crowding initially identified within a polyp were associated with neoplasia (EIN, carcinoma) on follow-up sampling. As the vast majority of repeat biopsies (91%) occurred within 12 months of the initial description of crowded glands, we suggest that this timeframe be the suggested clinical recommendation with the awareness that most follow-up sampling will occur within a 6- to 12-month timeframe. Overall, based on our series, it is our recommendation for 1 to 2 follow-up samples within a 6- to 12-month period. Consideration of continued sampling after a benign follow-up sample should be tempered by the clinical symptoms, as continued or renewed abnormal vaginal bleeding should prompt resampling in these patients. Common diagnostic terminology in our practice is: 'Crowded focus of cytologically altered glands, see Note. Note: Resampling is recommended in 6–12 months.'

It is of interest that no unifying morphological appearance of the cases descriptively diagnosed as 'gland crowding' could accurately predict future clinical outcome. This conundrum is identical to that faced in cervical cytology, and underscores both the importance of applying a set of clinically important criteria as well as the limitation imposed on predicting outcome when only a portion of the criteria are met.

In summary, this study shows that the term 'gland crowding' is useful in signifying interpretively difficult samples. The experience of others may differ, depending both on the classification used and the criteria applied. However, based on the results of this study, if some of the criteria for EIN are met, including gland crowding, nearly one-fourth could have a clinically significant outcome. Thus, we recommend a follow-up sampling within 6 months to 1 year.

Disclosure/conflict of interest

The authors declare no conflict of interest.

References

1 National Cancer Institute. Surveillance Epidemiology and End Results 2009, Available from http://seer.cancer.gov/statfacts/html/corp.html. Accessed on 22 June 2010.
2 Anastasiadis PG, Skaphida PG, Koutlaki NG, *et al.* Descriptive epidemiology of endometrial hyperplasia in patients with abnormal uterine bleeding. Eur J Gynaecol Oncol 2000;21:131–134.
3 Lidor A, Ismajovich B, Confino E, *et al.* Histopathological findings in 226 women with post-menopausal uterine bleeding. Acta Obstet Gynecol Scand 1986;65:41–43.
4 Montgomery BE, Daum GS, Dunton CJ. Endometrial hyperplasia: a review. Obstet Gynecol Surv 2004;59:368–378.
5 Clark TJ, Neelakantan D, Gupta JK. The management of endometrial hyperplasia: an evaluation of current

practice. Eur J Obstet Gynecol Reprod Biol 2006;125: 259–264.
6 Marsden DE, Hacker NF. Optimal management of endometrial hyperplasia. Best Pract Res Clin Obstet Gynaecol 2001;15:393–405.
7 McCluggage WG. My approach to the interpretation of endometrial biopsies and curettings. J Clin Pathol 2006;59:801–812.
8 Silverberg SG, Kurman RJ, Nogales F, *et al.* Tumors of the uterine corpus: epithelial tumors and related lesions In: Tavassoli FA, Devilee P (eds). WHO Classification of Tumors: Pathology and Genetics of Tumors of the Breast and Female Genital Organs. IARC Press: Lyon, France, 2003, pp 221–232.
9 Bergeron C, Nogales FF, Masseroli M, *et al.* A multicentric European study testing the reproducibility of the WHO classification of endometrial hyperplasia with a proposal of a simplified working classification for biopsy and curettage specimens. Am J Surg Pathol 1999;23:1102–1108.
10 Kendall BS, Ronnett BM, Isacson C, *et al.* Reproducibility of the diagnosis of endometrial hyperplasia, atypical hyperplasia, and well-differentiated carcinoma. Am J Surg Pathol 1998;22:1012–1019.
11 Mutter GL. Histopathology of genetically defined endometrial precancers. Int J Gynecol Pathol 2000;19:301–309.
12 Mutter GL, Baak JP, Crum CP, *et al.* Endometrial precancer diagnosis by histopathology, clonal analysis, and computerized morphometry. J Pathol 2000;190:462–469.
13 Baak JP, Mutter GL, Robboy S, *et al.* The molecular genetics and morphometry-based endometrial intraepithelial neoplasia classification system predicts disease progression in endometrial hyperplasia more accurately than the 1994 World Health Organization classification system. Cancer 2005;103:2304–2312.
14 Mutter GL, Kauderer J, Baak JP, *et al.* Biopsy histomorphometry predicts uterine myoinvasion by endometrial carcinoma: a Gynecologic Oncology Group study. Hum Pathol 2008;39:866–874.
15 Mutter GL, Zaino RJ, Baak JP, *et al.* Benign endometrial hyperplasia sequence and endometrial intraepithelial neoplasia. Int J Gynecol Pathol 2007;26:103–114.
16 Scully RE, Bonfiglio TA, Kurman RJ, *et al.* Uterine corpus. Histological Typing of Female Genital Tract Tumors. Springer-Verlag: New York, 1994, pp 13–31.
17 Zaino RJ. Endometrial hyperplasia: is it time for a quantum leap to a new classification? Int J Gynecol Pathol 2000;19:314–321.