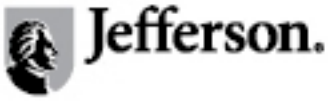

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Presence of endometrial adenocarcinoma in situ in complex atypical endometrial hyperplasia is associated with increased incidence of endometrial carcinoma in subsequent hysterectomy.

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Abstract:
The distinction of complex atypical endometrial hyperplasia from endometrial adenocarcinoma is often problematic. Foci of back-to-back arrangement of glands or foci of cribriform arrangement of glands smaller than 2.1 mm in diameter are considered insufficient for the diagnosis of endometrial adenocarcinoma by some authors, and sufficient to be diagnosed as endometrial adenocarcinoma by other authors. We refer to these foci as endometrial Adenocarcinoma-in-situ. In this study we evaluated findings in subsequent hysterectomy in complex atypical endometrial hyperplasia patients with and without Adenocarcinoma-in-situ. Follow-up findings, including the presence or absence of endometrial adenocarcinoma in the hysterectomy specimen, the grade of the carcinoma and the depth of myometrial invasion were analyzed. Of the total 87 patients with complex atypical endometrial hyperplasia, thirty-three patients had Adenocarcinoma-in-situ and 54 lacked Adenocarcinoma-in-situ. Twenty-two of 33 (66%) patients with Adenocarcinoma-in-situ had endometrial adenocarcinoma on subsequent hysterectomy versus 13 of 54 (24%) patients without Adenocarcinoma-in-situ (p= 0.0001). Myo-invasive endometrial adenocarcinoma was present in 20 of 33 (61%) patients with Adenocarcinoma-in-situ versus 8 of the 54 (15%) patients without Adenocarcinoma-in-situ (p=<0.0001). The depth of myometrial invasion in cases with myo-invasion was 24.5 + 19.4 % in patients with Adenocarcinoma-in-situ and12.8 + 8.5 % in patients without Adenocarcinoma-in-situ (p=0.05). Amongst patients younger than age of 50, 5 of the 7 (71%) with Adenocarcinoma-in-situ had myo-invasive carcinoma versus 2 of the 13 (15%) without Adenocarcinoma-in-situ (p=0.02). The likelihood of finding endometrial adenocarcinoma in subsequent hysterectomy in patients with complex atypical endometrial hyperplasia is significantly increased if Adenocarcinoma-in-situ is present in prior endometrial sampling. Endometrial adenocarcinomas in patients with Adenocarcinoma-in-situ are far more frequently myo-invasive, and invade to a greater depth than endometrial adenocarcinomas seen in patients without Adenocarcinoma-in-situ. Use of Adenocarcinoma-in-situ terminology could lead to improved management of patients with complex atypical endometrial hyperplasia.

Background: The distinction of complex atypical endometrial hyperplasia from endometrial adenocarcinoma is frequently problematic[1-11]. Foci of back-to-back arrangement of glands or foci of cribriform arrangement of glands smaller than 2.1 mm in diameter are considered insufficient for the diagnosis of endometrial adenocarcinoma by some authors[11], and sufficient to be diagnosed as endometrial adenocarcinoma by other authors[10]. We have referred to these foci as endometrial Adenocarcinoma-in-situ in the past. We have previously shown that the presence of such foci in complex atypical endometrial hyperplasia is associated with increased risk of finding endometrial carcinoma in subsequent hysterectomy[12]. In the current study we have used a larger number of cases of complex atypical endometrial hyperplasia from 3 different centers to further evaluate this association.

Design: Cases with the diagnosis of complex atypical endometrial hyperplasia on endometrial curettage/biopsy and subsequent hysterectomy were examined for the presence of Adenocarcinoma-in-situ. The cases were seen at New York University Medical Center, New York; North Broward Medical Center, Deerfield Beach, FL, and at Thomas Jefferson University Hospital, Philadelphia, PA. The cases were retrieved by searching for cases with the diagnosis of complex endometrial hyperplasia in the computerized records in each facility. The cases were sequential and seen from 2003 to 2006. Scant or otherwise sub-optimal specimens were excluded from the study. The presence or absence of Adenocarcinoma-in-situ was diagnosed at each facility based on review of the case by individual pathologists. Cytologic features were not included as a criteria for exclusion of a case, but none of the cases had grade III nuclei. The review of biopsies was done blindly without knowledge of the hysterectomy findings. Hysterectomy diagnosis of record was used.

Adenocarcinoma-in-situ was defined as foci of back to back arrangement of glands or foci of cribriform arrangement of glands composed of at least 4 glands and smaller than 2.1 mm in diameter (figs. 1 to 3). Foci with marked glandular crowding, where stromal cells were readily identified between adjacent glands, were not considered Adenocarcinoma-in-situ. Artifactual cribriform arrangement of glands like appearance can be seen when there is squamous metaplasia or morule formation in endometrial glands, and these were also not included as Adenocarcinoma-in-situ (fig. 4). The size of the largest Adenocarcinoma-in-situ focus was noted in each case. Follow-up findings in the two groups of patients with and without Adenocarcinoma-in-situ were analyzed, including the presence or absence of carcinoma in the hysterectomy specimen, the grade of the carcinoma and the depth of myometrial invasion.

Results: There were a total of 87 patients with complex atypical endometrial hyperplasia. The incidence of endometrial adenocarcinoma was 40% (35/87) and of myo-invasive carcinoma 32% (28/87) in subsequent hysterectomy in the entire group. All carcinomas were of endometrioid histology, and either grade I or II. Thirty-three patients (38%) had Adenocarcinoma-in-situ and 54 (62%) lacked Adenocarcinoma-in-situ. Twenty-two of 33 (66%) patients with Adenocarcinoma-in-situ had endometrial adenocarcinoma on subsequent hysterectomy versus 13 of 54 (24%) patients without Adenocarcinoma-in-situ

(p= 0.0001). Myo-invasive adenocarcinoma was present in 20 of 33 (61%) patients with Adenocarcinoma-in-situ versus 8 of the 54 (15%) patients without Adenocarcinoma-in-situ (p <0.0001). Myo-invasive carcinoma to a depth of 2 mm or more was present in 20 of the 33 (61%) patients with AIS, versus 6 of the 54 (11%) patients without Adenocarcinoma-in-situ (p <0.0001). Myo-invasive carcinoma to a depth of 3 mm or more was present in 15 of the 33 (45%) patients with AIS, versus 2 of the 54 (4%) patients without Adenocarcinoma-in-situ (p <0.0001). The depth of myometrial invasion in cases with myo-invasion was 24.5 $\pm$ 19.4 % in patients with Adenocarcinoma-in-situ and12.8 $\pm$ 8.5 % in patients without Adenocarcinoma-in-situ (p=0.05). The absolute depth of invasion in myo-invasive cases was 4.8 $\pm$ 3.5 mm in patients with Adenocarcinoma-in-situ and 2.1 $\pm$ 1 mm in patients without Adenocarcinoma-in-situ (p=0.01). A depth of invasion of greater than 50% was seen in 3 of the 33 patients with AIS, but in none of the 54 patients without Adenocarcinoma-in-situ (p=0.05). None of the carcinomas in either group were FIGO grade III. The larger size of the Adenocarcinoma-in-situ focus (>1 to <2.1 mm versus 1mm or smaller) was not predictive of subsequent carcinoma (p=0.68). The results are summarized in Table I.

Patients younger than age of 50

Amongst patients younger than age of 50, 5 of the 7 (71%) with Adenocarcinoma-in-situ had myo-invasive carcinoma versus 2 of the 14 (14%) without Adenocarcinoma-in-situ (p=0.02). In all 5 patients that had myo-invasive carcinoma in the Adenocarcinoma-in-situ group, the depth of invasion was 3 mm or greater. One of these patients had myo-invasion of 1 cm, equal to 58% of myometrial thickness. The two patients in the group without Adenocarcinoma-in-situ had 1mm and 2mm depth of invasion respectively. The results are summarized in Table II.

Discussion:

The overall incidence of endometrial adenocarcinoma and myo-invasive endometrial adenocarcinoma on follow-up hysterectomy in the group of complex atypical endometrial hyperplasia patients reported here was similar to what has been reported in the literature[6, 7, 11, 13, 14]. We found that the presence of Adenocarcinoma-in-situ in complex atypical endometrial hyperplasia patients was associated with significantly greater likelihood of finding endometrial adenocarcinoma and myo-invasive endometrial adenocarcinoma on subsequent hysterectomy. Approximately two thirds of patients with Adenocarcinoma-in-situ in endometrial curettage/biopsy have endometrial adenocarcinoma on subsequent hysterectomy versus about a quarter of those that lack Adenocarcinoma-in-situ. Endometrial adenocarcinomas in patients with Adenocarcinoma-in-situ are far more frequently myo-invasive, and invade to a greater depth than carcinomas seen in patients that have complex atypical endometrial hyperplasia without Adenocarcinoma-in-situ.

There is considerable confusion in the literature as to where complex atypical endometrial hyperplasia ends and endometrial adenocarcinoma starts. The distinction of endometrial carcinoma from complex endometrial hyperplasia has generally been based

on the criteria proposed by Kurman and Norris nearly 25 years ago[11, 15, 16]. In these studies, the cut off for endometrial carcinoma was arbitrarily set at 2.1 mm lesional size showing features of "Stromal invasion". However 7 of the 89 patients that lacked "Stromal invasion" also showed myo-invasive carcinoma in that study. It is unclear if any of these 7 patients with myoinvasive carcinoma had Adenocarcinoma-in-situ. This group without "Stromal invasion" included cases with complex atypical endometrial hyperplasia and "Carcinoma-in situ", but separate follow up data for patients in complex atypical endometrial hyperplasia and "Carcinoma-in-situ" groups was not provided[11]. In other words, no data was presented regarding the outcome of patients that showed smaller foci of what was called "Stromal invasion". Such lesions are diagnosed as complex atypical endometrial hyperplasia by some pathologists[11], as endometrial adenocarcinoma by others[10], and as endometrial adenocarcinoma can not be ruled out by yet others. King et al.[17] examined a group of patients that they called "adenocarcinoma without stromal invasion" and found endometrial carcinoma in 28% (12/43) and myo-invasive carcinoma in 16% (7/43) of these patients on follow up hysterectomy. Longacre at al.[18] have shown that glandular complexity captured by a pictorial architectural index, along with nuclear pleomorphism and prominence of the nucleoli are features most predictive for the presence of myo-invasive carcinoma in complex atypical endometrial hyperplasia. They also reported that extensive squamous differentiation and fibroblastic stroma do not contribute to prediction of myo-invasive endometrial carcinoma in subsequent hysterectomy. Hendrickson et al. did not find fibrous stroma in curettings from most patients with subsequent myo-invasive endometrial carcinoma[19].

We propose that foci of back to back glands or cribriform arrangement of glands smaller than 2.1 mm across be classified as Adenocarcinoma-in-situ. The term "Carcinoma-in-situ" was mistakenly applied to eosinophilic metaplasia of the endometrium many years ago[20] and is no longer used in that context. The concept that small foci of cribriform arrangement of glands be called "Carcinoma-in-situ" in the endometrium is not entirely new, and has been used in the past by Welch and Scully[21], by Vellios[22] and by Buehl et al[23]. This concept, however, was not strictly defined previously, its definition varied from author to author, and follow up data on these cases was not published. World Health Organization did not include any form of carcinoma in situ of endometrium in its classification[24] because of lack of agreement on its definition[21]. In the current study, we have provided a strict definition for Adenocarcinoma in-situ of the endometrium, and documented its prognostic significance.

The relationship between adenocarcinoma in situ diagnosed preoperatively and endometrial adenocarcinoma found in hysterectomy specimens is unclear. Some lesions might represent smaller foci of the same neoplasm whereas others might represent independent and incompletely developed, incipient invasive carcinomas or risk lesions. The following observations support the latter possibility in some cases. Multiple foci of Adenocarcinoma-in-situ can be seen without associated endometrial adenocarcinoma. Foci of Adenocarcinoma-in-situ are often fairly widely distributed, with intervening areas of complex atypical endometrial hyperplasia. We have also seen cases where the histo-morphologic appearance of these foci varies, suggesting their independent origin from each other (fig. 5).

Although we prefer to use the term Adenocarcinoma-in-situ, a number of alternative terms could be potentially used for this lesion. These would include terms such as CAH type I and II, CAH type A and B, endometrial adenocarcinoma without stromal invasion, minimal carcinoma, micro-carcinoma, micro-invasive carcinoma, CAH with focal glandular confluence and "microscopic focus of adenocarcinoma".

Adenocarcinoma-in-situ of endometrium should not be confused with Endometrial Intraepithelial Carcinoma (EIC)[25], which is an early form of uterine serous carcinoma characterized by growth of cells with high grade nuclei on the endometrial surface and in glands. These cells usually over-express p53. Cribriform arrangement of glands or back to back arrangement of glands is not seen in EIC. EIC usually arises in the background of an atrophic endometrium.

Recognition of the substantial risk of myometrial invasion following a diagnosis of adenocarcinoma in situ should allow gynecologists and patients to make better informed decisions when conservative, non-surgical management is considered. In the current study only 2 of the 14 patients with complex atypical endometrial hyperplasia without Adenocarcinoma-in-situ age 50 or under had myo-invasion, which was 1mm and 2 mm respectively. Two had carcinoma confined to the endometrium, while 10 had no carcinoma. Thus these patients can be managed conservatively. In contrast 5 of the 7 patients 50 or under with complex atypical endometrial hyperplasia with Adenocarcinoma-in-situ had myoinvasive carcinoma, and the depth of invasion in each of the 5 patients was 3 mm or more. One patient had 58% depth of invasion. Patients with complex atypical endometrial hyperplasia with Adenocarcinoma-in-situ may also be considered for lymph node sampling at hysterectomy if these lymph nodes are enlarged.

In Summary, presence of Adenocarcinoma-in-situ should be looked for and reported by pathologists in endometrial biopsies showing complex atypical endometrial hyperplasia because of a significantly greater likelihood of finding endometrial adenocarcinoma and myo-invasive endometrial adenocarcinoma in subsequent hysterectomy if Adenocarcinoma-in-situ is present.

References:

1. Allison KH, Reed SD, Voigt LF, Jordan CD, Newton KM, Garcia RL. Diagnosing Endometrial Hyperplasia: Why is it so Difficult to Agree? *Am J Surg Pathol*. 2008.
2. Hecht JL, Ince TA, Baak JP, Baker HE, Ogden MW, Mutter GL. Prediction of endometrial carcinoma by subjective endometrial intraepithelial neoplasia diagnosis. *Mod Pathol*. 2005;18:324-30.
3. 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;23:1102-8.
4. 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;22:1012-9.
5. Skov BG, Broholm H, Engel U et al. Comparison of the reproducibility of the WHO classifications of 1975 and 1994 of endometrial hyperplasia. *Int J Gynecol Pathol*. 1997;16:33-7.
6. Dunton CJ, Baak JP, Palazzo JP, van Diest PJ, McHugh M, Widra EA. Use of computerized morphometric analyses of endometrial hyperplasias in the prediction of coexistent cancer. *Am J Obstet Gynecol*. 1996;174:1518-21.
7. Zaino RJ, Kauderer J, Trimble CL et al. Reproducibility of the diagnosis of atypical endometrial hyperplasia: a Gynecologic Oncology Group study. *Cancer*. 2006;106:804-11.
8. Silverberg SG. Problems in the differential diagnosis of endometrial hyperplasia and carcinoma. *Mod Pathol*. 2000;13:309-27.
9. Soslow RA. Problems with the current diagnostic approach to complex atypical endometrial hyperplasia. *Cancer*. 2006;106:729-31.
10. Silverberg SG. Hyperplasia and carcinoma of the endometrium. *Semin Diagn Pathol*. 1988;5:135-53.
11. Kurman RJ, Norris HJ. Evaluation of criteria for distinguishing atypical endometrial hyperplasia from well-differentiated carcinoma. *Cancer*. 1982;49:2547-59.
12. Ventura KC, Popiolek D, Mittal K. Endometrial adenocarcinoma in situ in complex atypical hyperplasia: correlation with findings in subsequent hysterectomy specimen. *Int J Surg Pathol*. 2004;12:225-30.
13. Trimble CL, Kauderer J, Zaino R et al. Concurrent endometrial carcinoma in women with a biopsy diagnosis of atypical endometrial hyperplasia: a Gynecologic Oncology Group study. *Cancer*. 2006;106:812-9.
14. Widra EA, Dunton CJ, McHugh M, Palazzo JP. Endometrial hyperplasia and the risk of carcinoma. *Int J Gynecol Cancer*. 1995;5:233-235.
15. Kurman RJ, Kaminski PF, Norris HJ. The behavior of endometrial hyperplasia. A long-term study of "untreated" hyperplasia in 170 patients. *Cancer*. 1985;56:403-12.
16. Norris HJ, Tavassoli FA, Kurman RJ. Endometrial hyperplasia and carcinoma. Diagnostic considerations. *Am J Surg Pathol*. 1983;7:839-47.

17. King A, Seraj IM, Wagner RJ. Stromal invasion in endometrial adenocarcinoma. *Am J Obstet Gynecol*. 1984;149:10-4.

18. Longacre TA, Chung MH, Jensen DN, Hendrickson MR. Proposed criteria for the diagnosis of well-differentiated endometrial carcinoma. A diagnostic test for myoinvasion. *Am J Surg Pathol*. 1995;19:371-406.

19. Hendrickson MR, Ross JC, Kempson RL. Toward the development of morphologic criteria for well-differentiated adenocarcinoma of the endometrium. *Am J Surg Pathol*. 1983;7:819-38.

20. Hertig AT, Sommers SC, Bengloff H. Genesis of endometrial carcinoma III. Carcinoma in situ. Cancer 2:964-71, 1949.

21. Welch WR, Scully RE. Precancerous lesions of the endometrium. Hum Pathol 8:503-12, 1977.

22. Vellios F. Endometrial Hyperplasia and Carcinoma in-situ. *Gyn Oncol*. 1974;2:152-161.

23. Buehl IA, Vellios F, Carter JE, Huber CP. Carcinoma in situ of the endometrium. Am J Clin Path 42:594-601, 1964.

24. Poulson HE, Taylor CW. International Histological Classification of Tumours. No. 13. Histological Typing of Female Genital Tract Tumours. Geneva, World Health Organization, 1975.

25. Ambros RA, Sherman ME, Zahn CM, Bitterman P, Kurman RJ. Endometrial intraepithelial carcinoma: a distinctive lesion specifically associated with tumors displaying serous differentiation. *Hum Pathol*. 1995;26:1260-7.

Figure Legends:

Figs.1 to 3. Examples of Adenocarcinoma-in-situ in cases of complex atypical endometrial hyperplasia. 1a, 2a, and 3a, are lower power views (H & E, x 40). 1b, 2b, and 3b are the corresponding higher powers (H & E, x 200).

Fig.4. A cribriform like appearance can be seen due to squamous morule formation in endometrial glands. This should not be confused with Adenocarcinoma-in-situ (H & E, x 200).

Fig. 5. Occasionally, different foci of Adenocarcinoma-in-situ on the same slide have different morphologic features in the cells, suggesting their independent origin (H & E, x 200).

Table Legends:

Table 1.
Endometrial adenocarcinoma and myo-invasion was much more likely to be found in subsequent hysterectomy in cases with complex atypical endometrial hyperplasia on endometrial biopsy if Adenocarcinoma-in-situ was present in prior endometrial biopsy.

Table 2.
In a subgroup of patients younger than 50 years of age, Endometrial adenocarcinoma and myo-invasion was much more likely to be found in subsequent hysterectomy in cases with Adenocarcinoma-in-situ in complex atypical endometrial hyperplasia on endometrial biopsy.