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Review Article

Diagnostic and reporting issues of preneoplastic polyps of the large intestine with early carcinoma

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ABSTRACT

Premalignant polyps of the large intestine are common specimens in surgical pathology. They consist of several different subtypes identifiable by histological criteria that are associated with different molecular characteristics and with the development of different types of colorectal carcinoma. The most common of these is the conventional adenoma, which most commonly leads to carcinomas with a low degree of methylation (CIMP-L) that are microsatellite stable. In Lynch syndrome patients these polyps lead to CIMP-L carcinomas that are microsatellite instable. The second most common is the sessile serrated adenoma, which leads to carcinomas with a high degree of methylation (CIMP-H) that may be either microsatellite stable or instable. The least common premalignant polyp is the traditional serrated adenoma, which can lead to either CIMP-L or CIMP-H carcinomas, most often microsatellite stable. This paper will review the histological features of these lesions, discuss problems in diagnosis and discuss the role of histology in management.

It is generally accepted that most if not all carcinomas of the large intestine, excluding those arising in the background of chronic inflammatory bowel disease, develop in a preexisting polyp. For sporadic carcinomas, as well as carcinomas developing in familial adenomatous polyposis, MUTYH-associated polyposis, Lynch syndrome and serrated polyposis, these polyps are conventional adenomas (ConA), sessile serrated adenomas/polyps (SSA/P) and traditional serrated adenomas (TSA). Although some polyps generally considered hamartomatous such as juvenile retention polyps may also develop malignancy, this almost always occurs in the setting of a polyposis syndrome. The pathway to development of carcinoma and the types of resulting tumors varies depending on the precursor lesion, and may have implications for therapy [1,2]. ConAs most often produce microsatellite stable (MSS) CpG island methylator phenotype low (CIMP-L) carcinomas, although if the patient has Lynch syndrome conventional adenomas develop into microsatellite instable (MSI) CIMP-L carcinomas. About 60% of all colorectal carcinomas are MSS and CIMP-L. About 5% are MSI and CIMP-L and occur in Lynch syndrome. SSA/P develops into CpG island methylator phenotype high (CIMP-H) carcinomas that may be either MSS or MSI. This pathway accounts for approximately 30–35% of all sporadic carcinomas, as well as most carcinomas arising in serrated polyposis. The pathway to carcinoma development in TSA is still somewhat controversial although it appears that carcinomas arising via this pathway can be either CIMP-H or CIMP-L and most are MSS or show a low degree of microsatellite instability (MSI-L) [3]. This pathway probably accounts for < 5% of all sporadic carcinomas.

This article is intended to be a “primer” of diagnostic features and reporting suggestions for preneoplastic polyps of the large intestine and the carcinomas that develop in them. Details of the pathophysiology and molecular aspects of the various lesions will not be discussed except as necessary to explain the morphology.

1. Conventional adenoma

1.1. Diagnosis

Conventional adenomas arise from a mutation of the *APC* gene, which results in cellular dedifferentiation and unrestricted cellular division [4]. Hence, a required feature of these lesions should be mitotic activity at the surface of the polyp, away from the normal proliferative zone at the base of the crypts, in association with some degree of dedifferentiation of the cells (Figs. 1, 2). Although the textbook definition of a conventional adenoma defines adenoma by the presence of “dysplasia”, aside from increased mitotic activity, it is very difficult to find a clear definition of what the minimum criteria are for the identification of dysplasia. Typically dysplastic cells are less differentiated than normal colonic mucosa, having decreased mucin (although this is not always the case), and have nuclei that are considered “hyperchromatic”, which implies being more basophilic than normal nuclei (presumably due to increased chromatin due to chromosomal instability).

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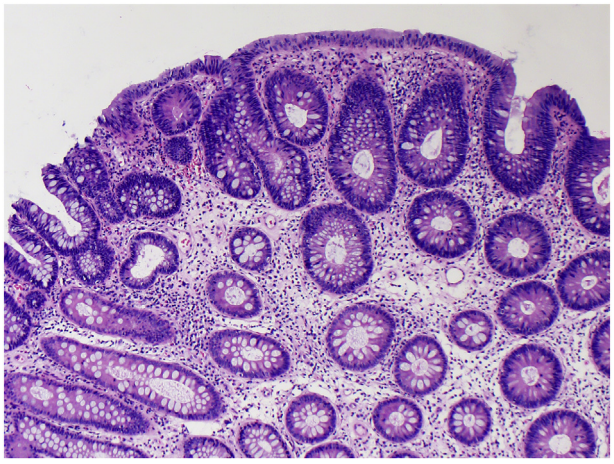

Fig. 1. Conventional tubular adenoma with low grade dysplasia. Note that the deeper crypts are not involved with dysplasia.

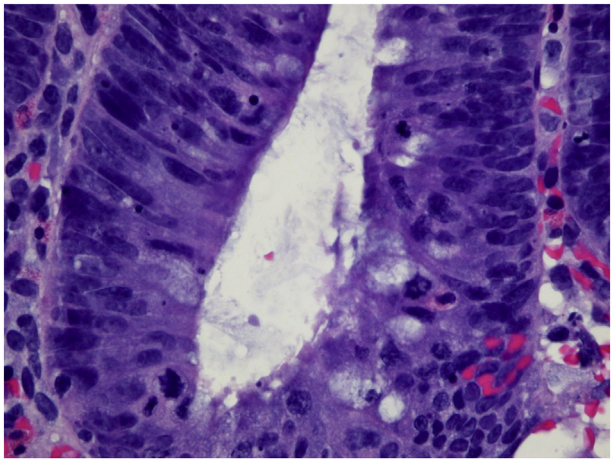

Fig. 2. Conventional tubular adenoma with low grade dysplasia. The dysplastic cells demonstrate elongated nuclei which appear stratified although the cells themselves are not stratified ("pseudostratified") and have uniform chromatin with small inconspicuous nucleoli. Mitoses can be identified, along with karyorrhexis from apoptotic cells.

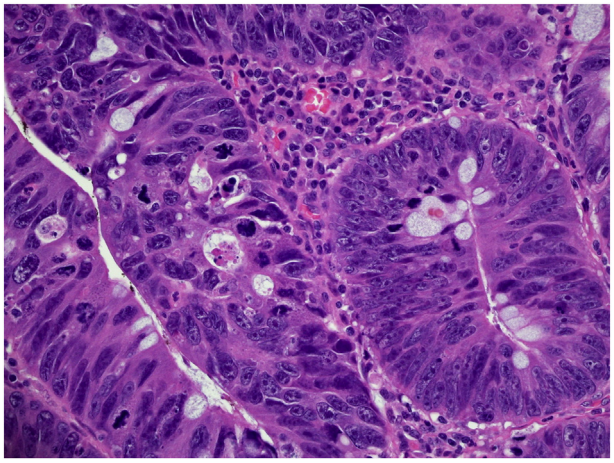

Fig. 3. Conventional tubular adenoma with high grade cytological dysplasia. In comparison with Fig. 2, many of the nuclei are rounded and demonstrate prominent single nucleoli. The cells appear truly stratified and mitoses are more numerous.

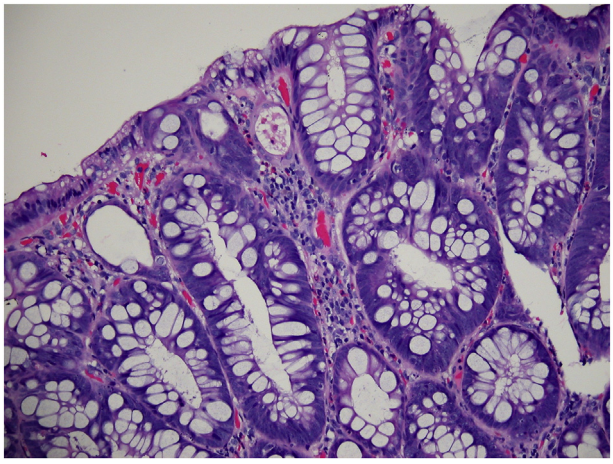

Fig. 4. Conventional tubular adenoma with abundant mucin. While most conventional adenomas have decreased mucin, in some cases goblet cells are abundant, as in this case. The diagnosis of tubular adenoma is based on the nuclear features of the cells and the presence of mitotic activity near the luminal aspects of the crypts.

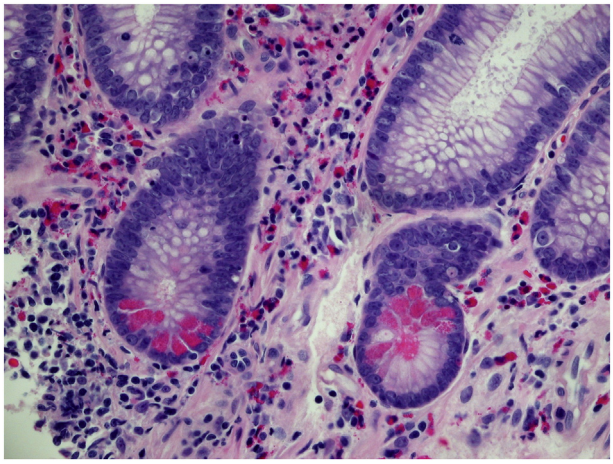

Fig. 5. Conventional tubular adenoma with prominent Paneth cell metaplasia. This is usually seen in the deeper aspects of the crypts.

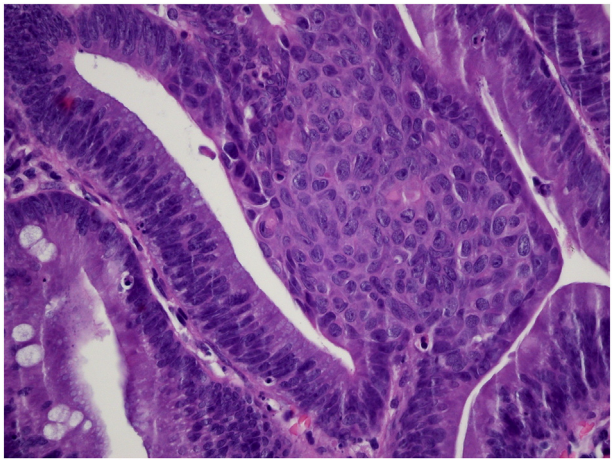

Fig. 6. Conventional tubular adenoma with morule formation. These squamoid-appearing cells with a moderate amount of eosinophilic cytoplasm can be seen near the base or mid portion of the crypts.

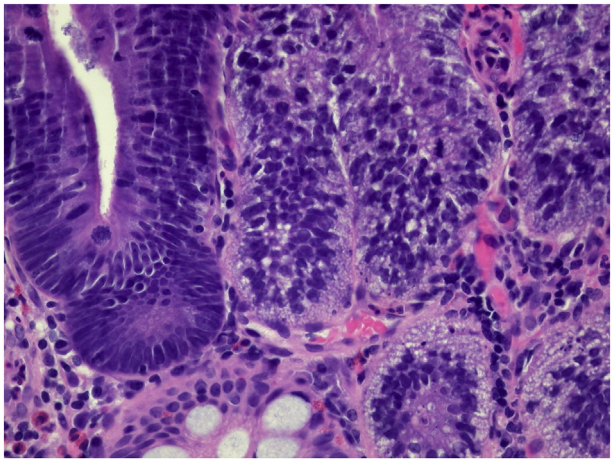

Fig. 7. Conventional tubular adenoma with clear cell change. There is a crypt showing conventional low grade dysplasia at the left, with the other crypts demonstrating bubbly clear appearing cytoplasm.

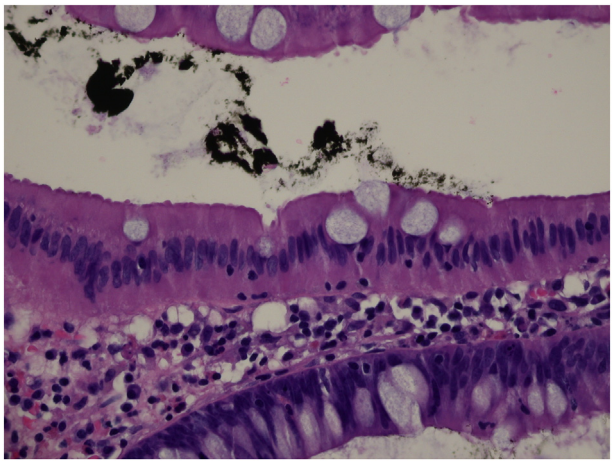

Fig. 8. Conventional tubular adenoma with enteric metaplasia. The crypt at the bottom demonstrates conventional low-grade dysplasia. The one near the center of the picture demonstrates cells with abundant eosinophilic cytoplasm, bland nuclei and an absence of mitotic activity.

Otherwise the shape and size of nuclei and the character of the cytoplasm has considerable variation from adenoma to adenoma. In typical low grade dysplasia the nuclei are elongated and pseudostratified with small inconspicuous nucleoli (Fig. 2). In higher grade conventional dysplasia the nuclei become round, may show true stratification and often have prominent nucleoli. In general, mitotic activity is greater in areas of high grade dysplasia (Fig. 3). By definition, dysplasia is found on the luminal aspects of conventional adenoma and may or may not involve the deep crypts (Fig. 1). Although in many conventional adenomas the cytoplasm of the tumor cells has little or no mucin, in some cases, particularly more villous tumors, there may be abundant intracytoplasmic mucin (Fig. 4). Several types of metaplasia may be superimposed on conventional adenomas including Paneth cell metaplasia (in about 20% of ConAs), morule formation (squamoid metaplasia)(0.4%), clear cell metaplasia (< 1%) and enteric metaplasia (currently unknown in frequency)(Figs. 5–8) [5-7]. While these various metaplasias may have some role in development of some rare malignancies of the colon (in particular, clear cell carcinoma from clear cell metaplasia and squamous carcinoma from squamous morules), the simple presence of these metaplasias does not play a role in management of ConAs.

In addition to cytological variation, there is considerable architectural variation in the overall growth pattern of the neoplastic cells.

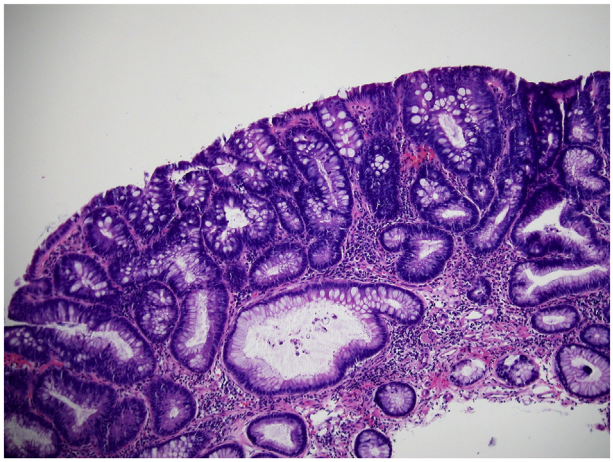

Fig. 9. Conventional tubular adenoma with low-grade dysplasia and with a much more complex growth pattern than that seen in Fig. 1.

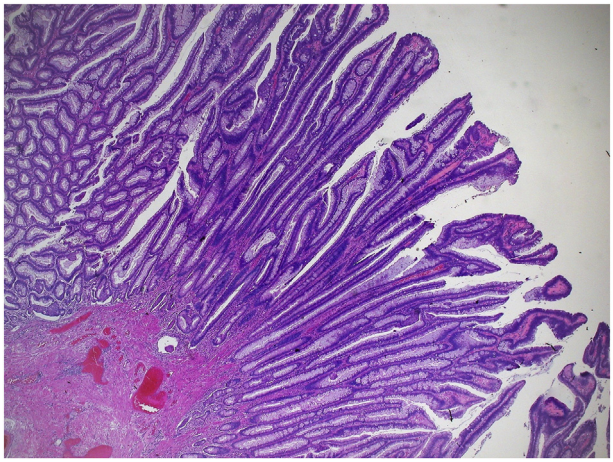

Fig. 10. Conventional villous adenoma. In this lesion the low grade dysplastic epithelium is located on the outside surface of long finger-like projections of lamina propria.

They may have the appearance of simple growth of dysplastic cells in normal preexisting crypts (Fig. 1), or may consist of more complex arrays of tubules the same size or smaller than normal crypts (Fig. 9). ConAs have also have varying proportions of villous growth (Fig. 10). It seems probable that these variations in growth pattern reflect molecular differences that have yet to be well defined aside from a reported association between *KRAS* mutation and villous architecture [8,9].

1.2. Classification of ConA

Conventional adenomas are generally categorized by overall growth pattern into tubular (TA), tubulovillous (TVA) and villous (VA) adenomas and by the degree of cytological dysplasia into those with and without high grade dysplasia. Villi are defined as long fingerlike projections with the neoplastic cells being present on the outer surface of the villi, rather than being present within crypt-like structures resembling normal crypts (Fig. 10). It has been well demonstrated that lesions with more villous architecture are more likely to harbor carcinoma at the time of their detection and hence they are considered closer to carcinoma along the adenoma – carcinoma sequence of Fearon and Vogelstein [10]. Therefore, these lesions have been termed “advanced adenoma” (i.e. adenomas more advanced toward carcinoma). It is likely that the increased surface area generated by villous architecture leads to a greater propensity to develop secondary mutations due to exposure

of the proliferating population of cells to promoters present in the stool. The division into villous subtypes is arbitrarily defined by the percentage of villous component. Typically, < 25% villous is considered a TA, 25–50% TVA and over 50% VA. Many years ago there was considerable debate regarding the possibility that any villous component, regardless of amount, might be important (and in point of fact, there were heated arguments during that time with some authors contending that pure tubular adenomas had no or negligible malignant potential), and recent data would suggest that in point of fact, even small areas of villous architecture might be important, increasing the risk of malignancy [11].

ConAs are also classified on the basis of the degree of dysplasia into those with only low grade dysplasia and those with high grade dysplasia. High grade dysplasia is a feature found more commonly in ConAs harboring carcinoma and hence is another feature of "advanced" adenoma. High grade dysplasia is perhaps more difficult to define than villous architecture. Generally both cytological and architectural features are taken into account. Cytologically high grade dysplasia is characterized by round somewhat pleomorphic nuclei with prominent nucleoli, often demonstrating a higher mitotic rate than the surrounding low grade dysplasia (Fig. 3). The cells demonstrate true stratification rather than pseudostratification, and this excess growth eventually leads to architectural complexity with the formation of back to back glands (Fig. 11).

1.3. Problems with the diagnosis of ConA

Issues with ConA occur in relationship to diagnosis (i.e. to identification of the lesion) and more commonly in regard to subclassification.

Most ConAs are very straightforward to diagnosis. Most underdiagnoses relate to insufficient sectioning of the tissue. Studies have shown that in at least 10% of cases, when the endoscopist sees a polyp and there is no polyp on the initial histological sections, cutting additional levels will identify a lesion that was missed [12]. This is no doubt simply due to the orientation of a small biopsy specimen in the paraffin block. Occasional careful examination will reveal a single small crypt with cytological dysplasia that will become more apparent on deeper levels. Occasionally ConAs will have accompanying mucosal hyperplasia that will resemble a goblet cell rich hyperplastic polyp (Fig. 12). Therefore, when there are changes in a biopsy that are marginally diagnostic of a GCHP additional levels may reveal a ConA. Overdiagnosis of ConA can occur when there is reactive atypia at the edge of an ulcer that resembles ConA. In general caution is recommended when dealing with ulcerated lesions, since most ConAs do not ulcerate, or if they do then one should worry about possible malignancy. Reactive epithelial changes at the edge of an ulcer can look very much like ConA but can

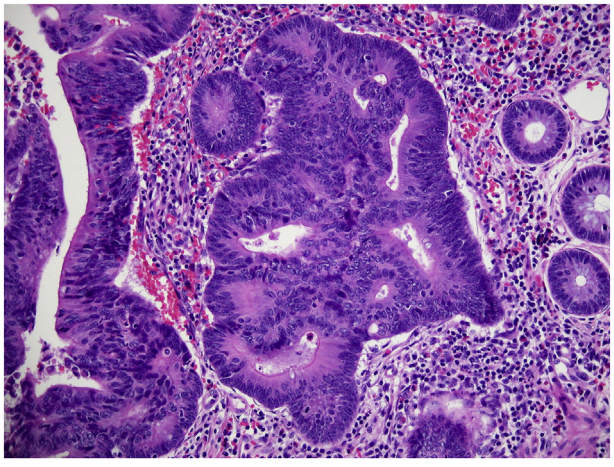

Fig. 11. Conventional tubular adenoma with architectural high grade dysplasia. The tumor cells are arranged in a complex arrangement with multiple lumina created by bridges of cells unsupported by stroma. In this particular example the cytology is not particularly high grade.

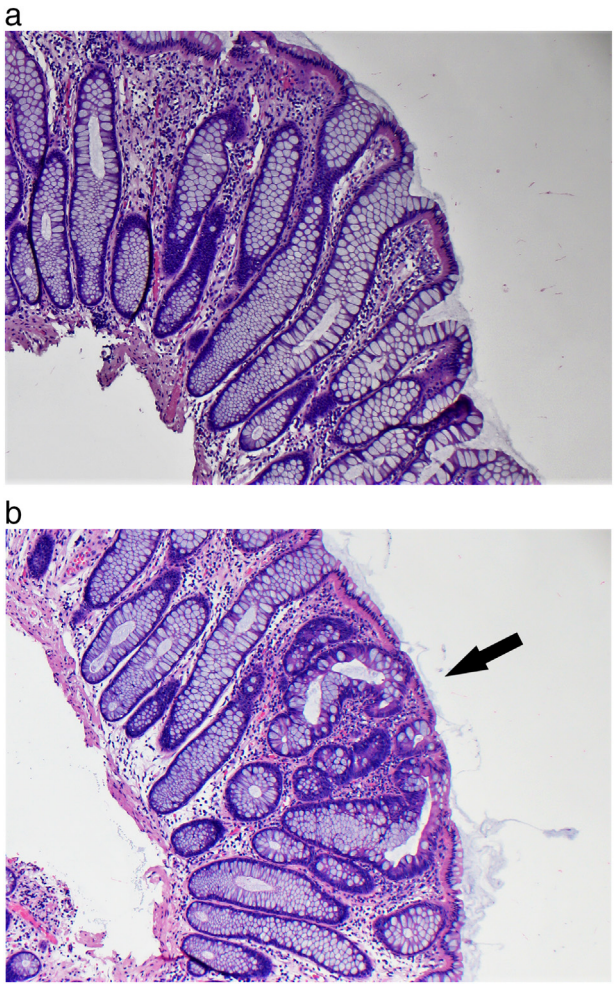

Fig. 12. A. The first 3 tissue levels of a rectal polyp demonstrating increased goblet cells, crypt elongation and subtle serration suggestive of goblet cell rich hyperplastic polyp. B. Deeper levels demonstrated a small tubular adenoma in the center of the lesion (arrow).

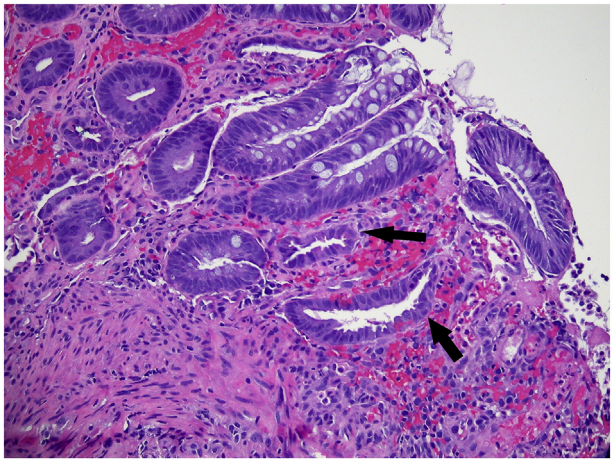

Fig. 13. Rectal biopsy from an erosion caused by a rectal stent. Although much of the biopsy appears "dysplastic", the right side of the biopsy demonstrates an erosion and there are several clearly reactive crypts in close association with the more dysplastic area (arrows). This is all considered reactive atypia.

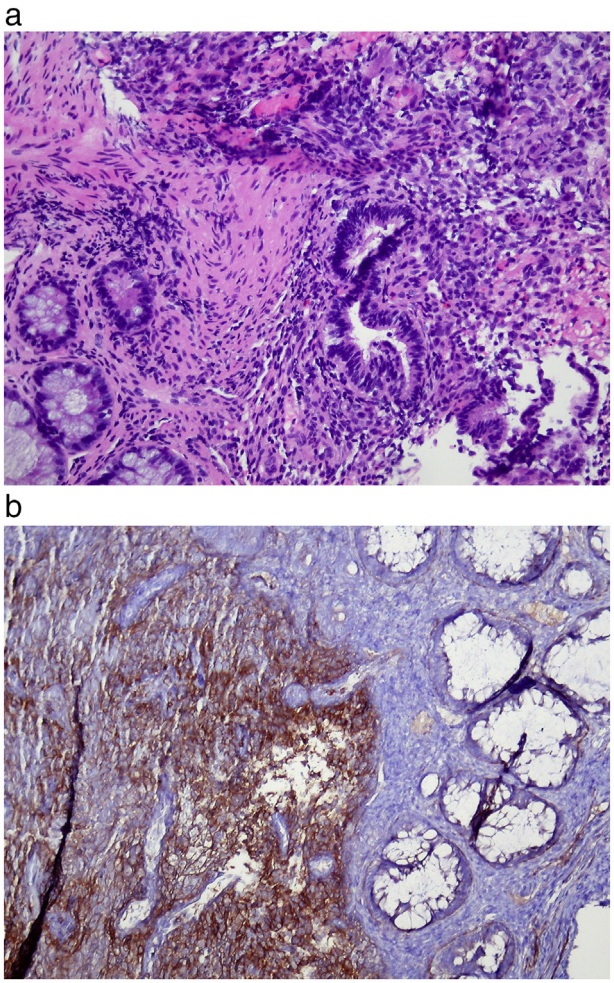

Fig. 14. A. Biopsy of a nodule in the rectum. Note the presence of glands that appear dysplastic near the center of the biopsy. This area is suggestive of adenoma but the stroma is hypercellular and does not look like typical lamina propria. B. A CD10 stain confirms the presence of endometrial stroma consistent with endometriosis.

usually be distinguished by the presence of clearly reactive epithelium in direct continuity with the more dysplastic appearing cells (Fig. 13). Since ConAs are clonal processes, there should be a clear and sharp demarcation between the adenoma and the adjacent normal mucosa, with no transitional-appearing cells. A rare condition that can simulate ConA is endometriosis involving the colon (Fig. 14), which can usually be recognized by the accompanying endometrial stroma. If one thinks about this possibility, staining for CK7 and 20 (endometrosis with be CK7+, CK20- and ConA the reverse) as well as CD10 to identify the stroma will solve the problem.

In regard to classification, neither villous architecture nor high grade dysplasia has a high degree of reproducibility. In the better designed reproducibility studies (i.e. those studies looking at consecutive series of unselected specimens), the kappa statistic for both changes is generally poor, in the range of 0.30–0.35 [13,14]. In regard to villous architecture, defining what constitutes a "villous structure" is not easy. Some adenomas have short "stumpy" villous structures lining the surface (Fig. 15), whereas other lesions have long villi that extend from the muscularis mucosae into the bowel lumen (Fig. 10). Some pathologists would consider the former to constitute villi, whereas others would not, and at this point there are no compelling data to determine which is the correct interpretation. In addition, it is not clear what the "25%" figure required for moving a TA to TVA refers to. Is it the surface area or the volume of the adenoma? And how does one actually determine the

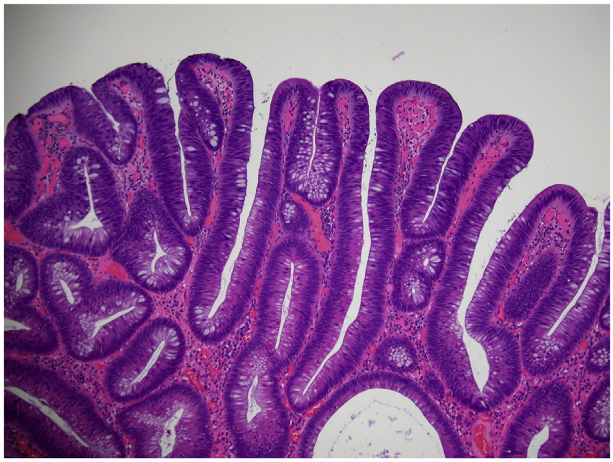

Fig. 15. Conventional adenoma with short villous projections on the surface. Cases such as these are often difficult to classify and are a cause of interobserver disagreement.

percentage of a three dimensional polyp when viewed in only 2 dimensions? In addition, as noted above, recent data would suggest that any villous architecture is important to outcome, not just 25% villous architecture [11]. Reproducibility of high grade dysplasia is even worse, even in studies where pathologists do training to improve results. This is even more difficult to explain, since most pathologists equate HGD with "carcinoma in situ" which one would think is generally easy to diagnose. One would think that a CIS-like degree of dysplasia would be reproducible but tangential sectioning can produce a false impression of stratification or architectural complexity, which often seems to be over interpreted as high grade dysplasia. One would hope that only diagnosing HGD when both architectural complexity and cytological high grade changes are present would alleviate some of the discrepancy. Regardless of the reasons, the fact that these features are not reproducible casts significant doubt on their significance, particularly in regard to determining rescreening intervals [15]. For this reason, some authors have recommended discontinuing subclassification of conventional adenomas [16]. While in general I intellectually agree with this sentiment, essentially all screening guidelines currently require the degree of villous architecture and high grade dysplasia to be used for rescreening decisions (despite a lack of strong data supporting this recommendation), therefore as a practical matter pathologists should still make a good faith effort to put these data into their reports [17].

2. Carcinoma arising in conventional adenomas

2.1. Diagnosis

Most carcinomas arising in conventional adenomas are adenocarcinoma, although high grade neuroendocrine carcinomas, squamous cell carcinoma and clear cell carcinoma also rarely are seen arising in these lesions [18-20]. Most discussion regarding management of carcinomas in ConA refers to adenocarcinoma, however. This is particularly true given that these rare forms of carcinoma typically have a very bad prognosis and are rarely identified while still confined to the polyp.

Essentially all adenocarcinomas arising in ConA are found in association with and presumably arise from high grade dysplasia. For many years no distinction was drawn between high grade dysplasia and intramucosal carcinoma (i.e. carcinoma invading into the lamina propria but no deeper), however this distinction is of some clinical importance and has now been recognized in the 8th edition of the AJCC staging manual [21]. High grade dysplasia without invasion into the lamina propria is now classified only as HGD and is not included in the

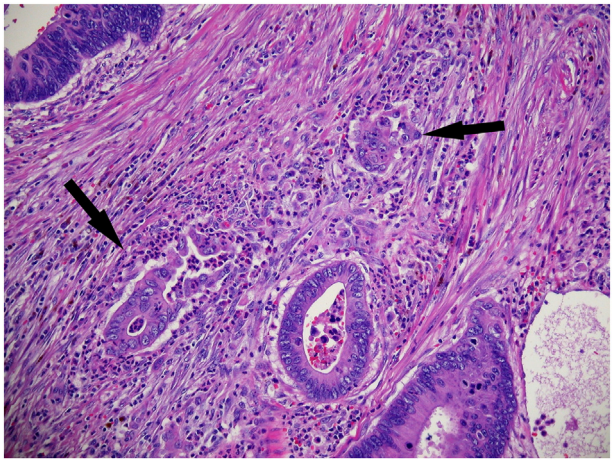

Fig. 16. Intramucosal carcinoma in a tubular adenoma. Note the presence of variably sized infiltrating glands (arrows) and the desmoplastic stroma.

category of Tis as had been done in prior editions of AJCC. Although it seems somewhat oxymoronic, "Tis" now refers only to tumors with invasion into the lamina propria or into but not through the muscularis mucosae (Figs. 16 and 17). This is important because, contrary to much early teaching, lymphatic vessels are present in the lamina propria of normal colon and are even more prevalent in neoplastic polyps [22]. Therefore, tumors invasive only into the lamina propria do have access to lymphatics and on rare occasion do result in the development of lymph node metastases (Fig. 17). A paradox of the current AJCC classification is that, despite the fact that intramucosal carcinoma is recognized as invasive, it is still stated that tumors are only considered "invasive", and therefore receive a designation of "T1", when they have invaded through the muscularis mucosae into the submucosa (Fig. 18). The diagnosis of adenocarcinoma arising in ConA is generally not problematic if the tumor is invasive through the muscularis mucosae (T1), and the usual criteria for malignancy apply. The distinction of intramucosal carcinoma from HGD can be more problematic.

2.2. Problems in diagnosis of adenocarcinoma in ConA

As noted above, identification of tumor invasion through the muscularis mucosae in ConA is usually not problematic. When neoplastic glands are seen below the muscularis mucosae the only differential diagnosis is so-called pseudoinvasion of the stalk [23] (Fig. 19). Pseudoinvasion is thought to result from damage to the adenoma, usually by torsion, with the creation of ulceration and displacement of adenoma cells into the submucosa. It is usually recognized by the presence of pools of mucin with or without neoplastic cells, surrounded by a cellular stroma usually with chronic inflammation and almost invariably with hemosiderin-laden macrophages indicative of the prior injury (Fig. 20). The tumor cells typically demonstrate only low grade dysplasia and are not worrisome for malignancy on a cytological basis when there is no HGD in the overlying adenoma. If pseudoinvasion occurs in adenomas with high grade dysplasia, however, and if the HGD is present in the pseudoinvasion, absolute diagnosis can be difficult if not impossible. In some cases use of cytokeratin staining to look for individual invasive cells can be of value.

Occasionally biopsy of a lesion will result in mechanical displacement of small clusters of neoplastic cells into the submucosa. These are different from pseudoinvasion in that mucin pools are usually not present nor is there hemosiderin deposition. Recognition that the lesion had been previously biopsied is reassuring under these circumstances [24].

Distinction of HGD from intramucosal carcinoma can be more problematic than identifying carcinoma in the submucosa. HGD is

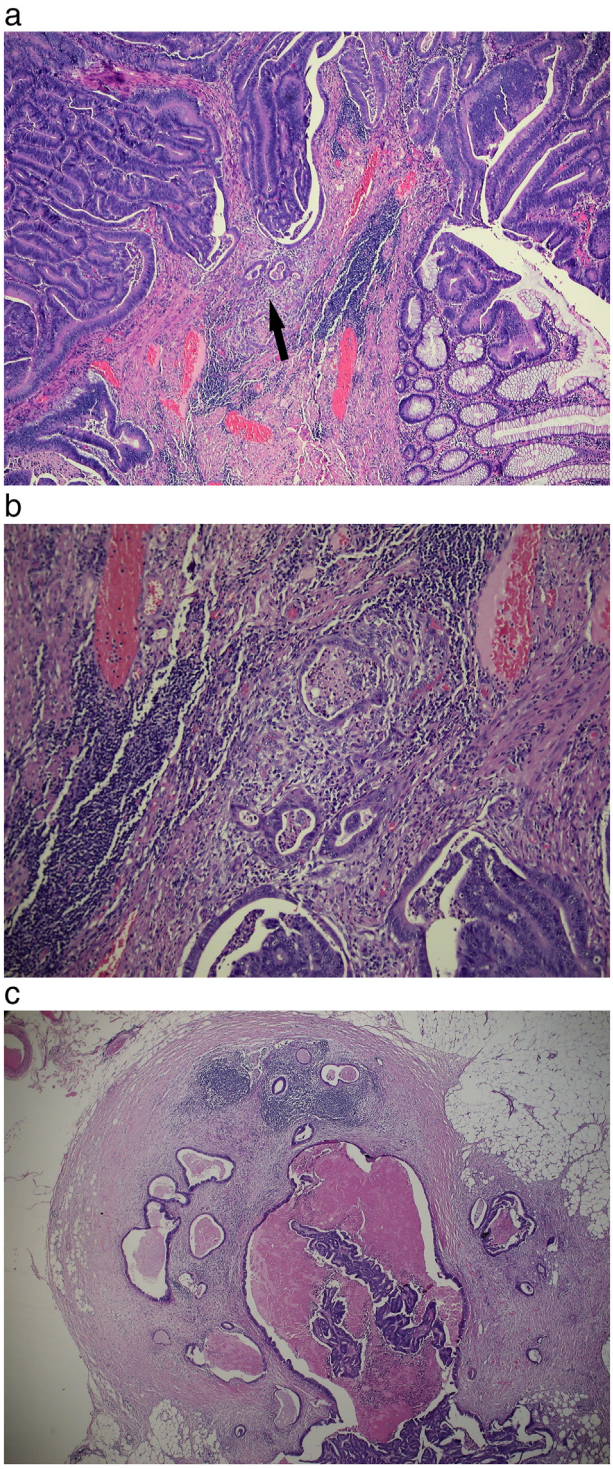

Fig. 17. A. Tubular adenoma with extensive high grade dysplasia and several infiltrative glands at the base of the polyp (arrow) B. Higher power of the area noted in A. The glands are infiltrate into the muscularis mucosae but do not invade into the submucosa (Tis). (C) Despite the absence of submucosal invasion there was evidence of lymphatic invasion and there were several lymph nodes positive at the time of resection.

defined as being confined within the basement membrane of crypts, although extensive cribriform growth with expansion of the crypts can sometimes cause difficulties. Usually if there is intramucosal invasion there is a desmoplastic reaction in the lamina propria and one can also identify small clusters of cells that are cytologically distinct from the HGD, often having no lumen and growing in an infiltrative pattern

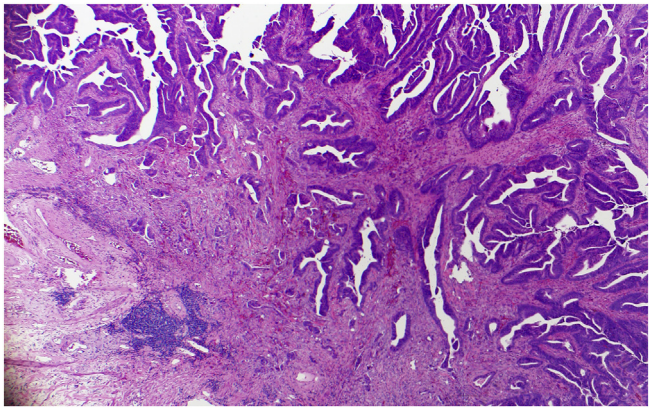

Fig. 18. Tubulovillous adenoma with early carcinoma invasive into the submucosa.

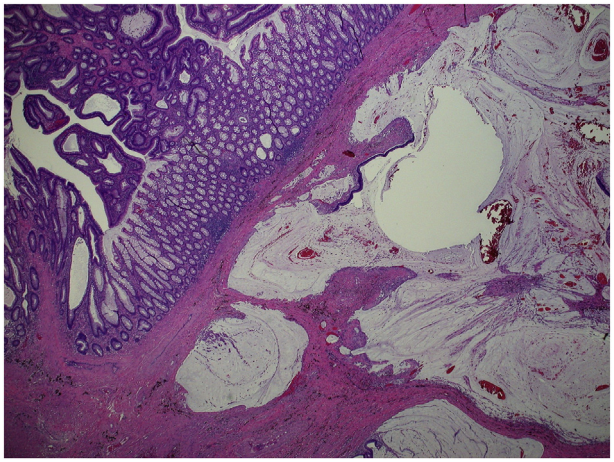

Fig. 19. Tubulovillous adenoma with several nests of mucin and epithelial cells in the submucosa characteristic of "pseudoinvasion", or displaced glands.

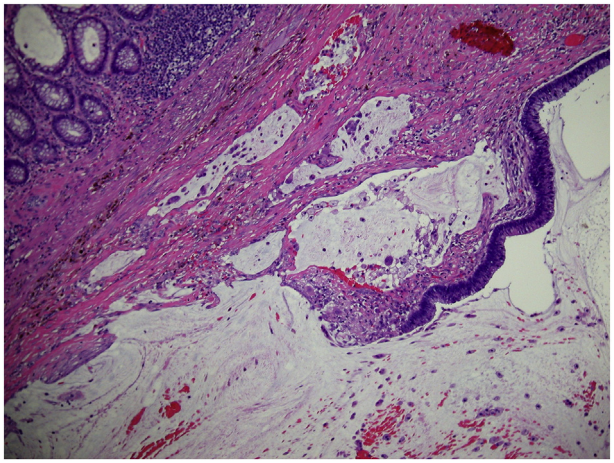

Fig. 20. In areas of pseudoinvasion the mucinous nests can be partially lined with low grade dysplastic epithelium and the surrounding stroma has hemosiderin deposition and chronic inflammation indicative of prior injury.

(Fig. 16). As with pseudoinvasion, the use of a cytokeratin stain may allow identification of these small clusters or of single cells, hence allowing the diagnosis (Figs. 21, 22).

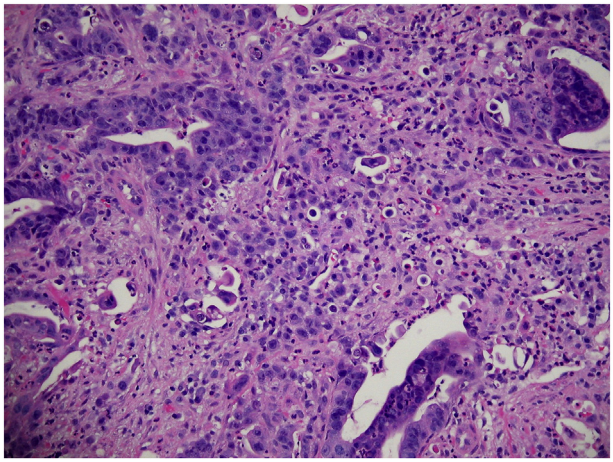

Fig. 21. Tubular adenoma with intramucosal carcinoma with a high degree of tumor budding. Large numbers of invasive single cells and clusters of cells can be seen between the dysplastic glands.

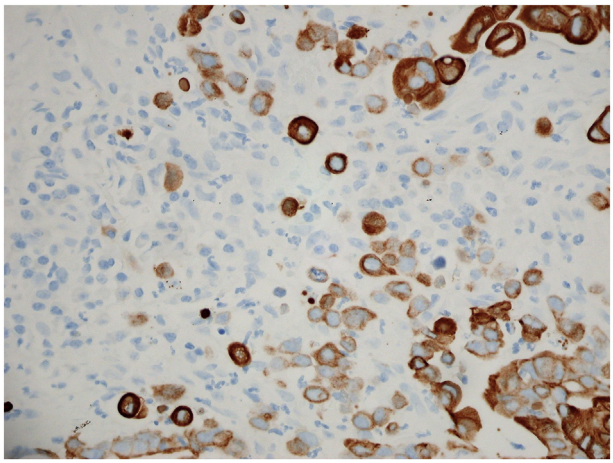

Fig. 22. Cytokeratin AE1/AE3 stain of the polyp in Fig. 21. The stain highlights large numbers of single invasive cells (tumor buds).

2.3. Histology in the management of carcinoma arising in ConA

Once carcinoma is identified in a ConA, histology may play a role in determining the proper management of the lesion. If the adenoma was simply biopsied or was resected piecemeal without a clear resection margin, then additional resection of the lesion is generally necessary. Grading the tumor and looking for tumor budding and lymphovascular invasion can be important in guiding therapy, even in these partially resected cases, since the options include immediate reexcision of the area of the lesion (if there is a reasonable chance based on the initial endoscopic procedure that the lesion was not completely resected) versus reendoscopy after some interval of time to see if the lesion has regrown (if the lesion was thought to be completely resected at the initial visit).

For lesions that are completely resected intact at the initial endoscopy, a number of features are used to predict risk of recurrence and of lymph node metastases.

The major factor predicting local recurrence is the status of the resection margin. Although technically this refers to the carcinoma margin more than the adenoma, the presence of residual adenoma at the margin should also be mentioned since benign adenoma at the margin may indicate a need for localized reexcision of the area or very close followup. In terms of the carcinoma, any carcinoma at the margin or within 1 mm of the margin is associated with a high risk of

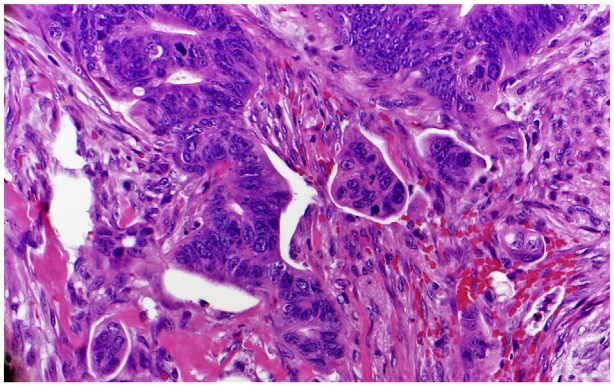

Fig. 23. Invasive micropapillary carcinoma in a tubulovillous adenoma. High power illustration of the invasive tumor Fig. 18 demonstrates small clusters of invasive cells within small spaces. The cells in these clusters appear to have their "luminal" surface oriented toward the periphery of the cluster in a fashion characteristic of micropapillary carcinoma.

recurrence [25]. Therefore a positive margin is considered tumor at the margin or within 1 mm of the margin. Ideally the margin will be identified in the gross room and inked for identification, but cautery artifact can also be used to identify the margin.

In regard to predicting lymph node metastases, several factors are considered important. These include grade of tumor, presence of lymphovascular invasion, presence of a high degree of tumor budding and the depth of invasion of the carcinoma [26].

Regarding grade of tumor, in general this is of relatively little practical value. The vast majority of carcinoma arising in ConA are well or moderately differentiated adenocarcinoma NOS. Only "poorly differentiated" tumors are associated with a higher risk of metastases. This would by definition include tumors with < 50% glandular formation, signet ring carcinoma, microsatellite stable mucinous carcinomas and high grade neuroendocrine tumors. All of these subtypes together probably account for < 5% of early invasive carcinomas seen. Micropapillary carcinoma can also arise in adenomas and may be more common than the above subtypes although it has not been extensively reported [27] (Figs. 16 and 23). Based on the known aggressive behavior of micropapillary carcinoma especially in regard to lymph node metastases, this tumor type should probably be considered a high risk tumor type when seen in adenomas.

Lymphovascular invasion predicts metastases and should be carefully evaluated. Artifactual clefting and a micropapillary growth pattern can simulate invasion, therefore use of a D2–40 or ERG stain to identify lymphatics can be of value in suspicious cases. ERG may be a more sensitive marker since it marks venous and arterial endothelium as well as lymphatic endothelium, and because cross reactivity may be less of an issue, although since it is a nuclear stain I find it somewhat harder to use to assess tumor in vascular lumena [28].

Tumor budding is now recognized as having considerable predictive value for lymph node metastases. Tumor budding is defined as invasive clusters of 5 or fewer tumor cells, usually evaluated at the leading edge of resected tumors although it has been demonstrated that intratumoral tumor budding is also significant. The currently recommended method for assessment is to count the number of tumor buds in a single 0.785 mm^2 field (generally a 20× objective) in the most active part of the tumor. > 10 buds is considered a high degree of tumor budding (Figs. 21, 22).

Finally, assessment of depth of invasion is well recognized as a predictor of metastases. There are several methods of assessment in the literature. One of the oldest and best known is the Haggitt system which classifies the depth of invasion in to 4 categories based on anatomic subdivisions [29]. The system is mainly useful for pedunculated polyps removed intact, and therefore is applicable to only a small fraction of

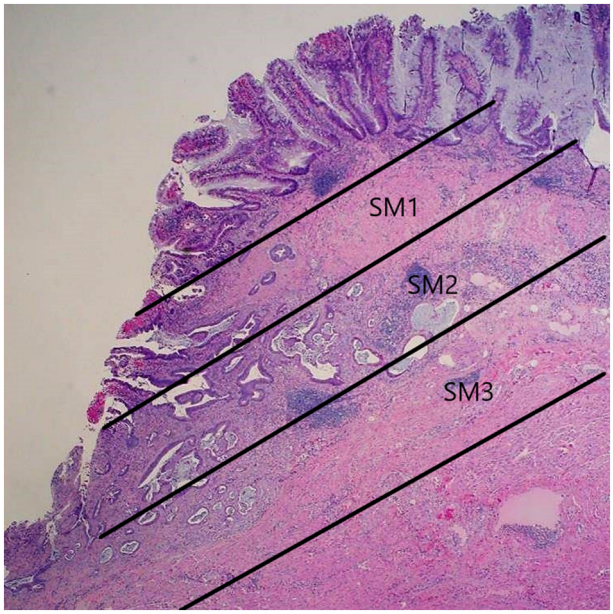

Fig. 24. Tubular adenoma with adenocarcinoma invasive into the submucosa. This illustrates subdivisions of the submucosa into thirds (SM1, SM2 and SM3). This system cannot be used unless muscularis propria is included in the specimen.

sampled specimens. The categories are also relatively crude with true clinical significance only for Haggitt level IV. A second, commonly used method is that of Kikuchi [30]. This system subdivides the submucosa into thirds and assigns categories of SM1, SM2 and SM3 for tumors invasive into the superficial, middle and deepest third of the submucosa (Fig. 24). The system shows a significant increase in the risk of lymph node metastases for SM3 (23%) versus SM1 + SM2 (3% and 8%). However, in order to use this system the entire depth of the submucosa must be included in the resection (i.e. the resection needs to include at least the superficial portion of the muscularis propria), which limits its applicability. Finally, a simple measurement of the depth of invasion beneath the muscularis mucosae has been shown to be of significance [31]. The risk of lymph node metastases is lower with invasion < 1 mm, and increases significantly with deeper invasion. Of the three systems, the simple submucosal measurement is the most generally applicable and easiest to apply to most specimens and is the one that I would recommend in all cases.

3. Sessile serrated adenoma/polyp

3.1. Diagnosis

Sessile serrated adenomas/polyps (SSA/P) are characterized by an overall serrated configuration and generally a flat or sessile growth pattern although rare large examples do occasionally appear pedunculated. Mechanistically it appears that SSA/P develop because of decreased cell death associated with activating mutations of the *BRAF* gene (a feature in common with the microvesicular type of hyperplastic polyp), and movement of the proliferation zone away from its usual location at the base of the crypts [1,32]. The decreased cell death results in an expanded population of mature or hypermature cells that accumulate creating the serrated appearance (Fig. 25). The movement of the proliferative zone results in bidirectional maturation of cells both toward the surface in a normal fashion, and toward the base of the crypts, resulting in abnormal basal maturation and in the formation of bizarre complex crypt shapes often referred to as boot or anchor shaped (Figs. 25 and 26). This resulting abnormal growth is often referred to as crypt distortion and is the one criterion most often used for the

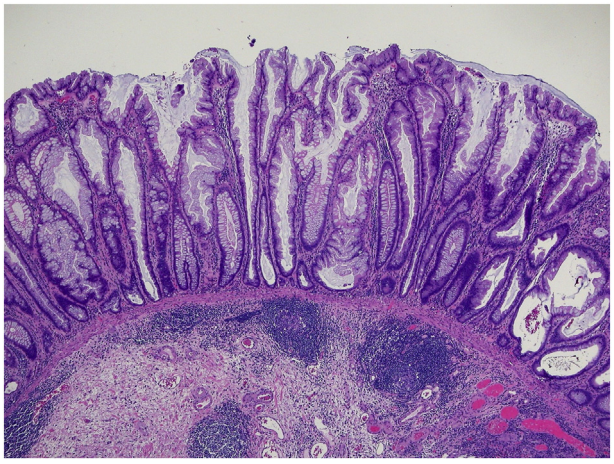

Fig. 25. Sessile serrated adenoma. The lesion appears hyperserrated. Notice that in addition to abnormally shaped crypts (also see Fig. 26), the entire lesion is excessively complex with serrations that are present deeply in the crypts rather than just at the surface, and there are many dilated crypts associated with excess mucin production.

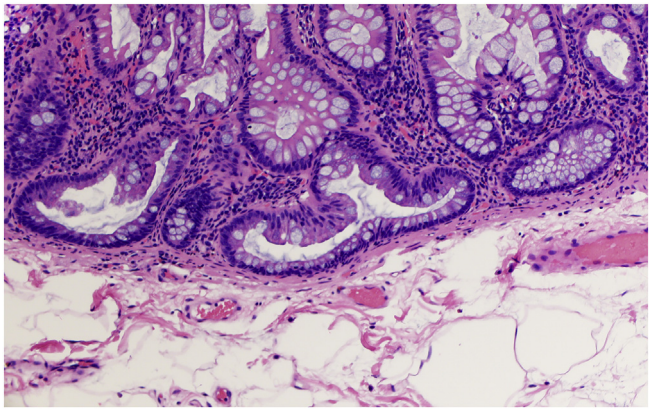

Fig. 26. Sessile serrated adenoma. These are "classic" anchor shaped crypts showing lateral growth along the muscularis mucosae. Note that these crypts are lined by abundant goblet cells.

diagnosis of SSA/P. This downward growth also results in herniation of epithelium through the muscularis mucosae in areas of vascular penetration, creating a so-called "inverted" growth pattern in which nests of mature epithelial cells are seen beneath the muscularis mucosae. Although distorted crypts are the most obvious and easily identified criterion for SSA/P, other features such as an overall more exuberant growth pattern (compared to HPs), the presence of marked variation in the size of the serrated crypts, and the presence of excess serration deep in the crypts (even without boot or anchor-shaped crypts) are useful diagnostic criteria (Fig. 25). In SSA/P not all crypts are distorted and many areas resemble hyperplastic polyps, so only a portion of the lesion needs abnormal crypt growth to be considered SSA/P. The number of abnormal crypts reportedly needed for the diagnosis has been arbitrarily defined in different publications from 1 to 3, however this is an oversimplification of the diagnosis and other features as mentioned above can be used to support the diagnosis in cases of uncertainty. In the majority of cases reliance on a single abnormal crypt is not necessary for diagnosis.

As noted in the introduction, SSA/P can progress to carcinoma of several molecular types. In general carcinomas arising from these lesions are *BRAF* mutated (about 80% have a mutation at the V600E locus) and show a high degree of methylation (CIMP-H). They can be microsatellite stable or instable. The first sign of progression to

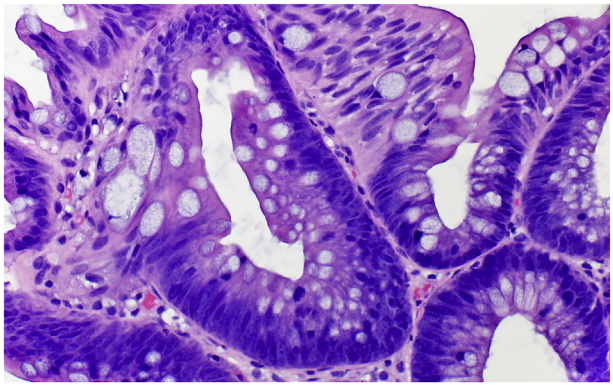

Fig. 27. Sessile serrated adenoma with conventional cytological dysplasia. Notice the abrupt transition from bland serrated epithelium to pseudostratified mitotically active epithelium.

carcinoma is the development of frank cytological dysplasia in the SSA/P, at which point the lesion is diagnosed as SSA/P with cytological dysplasia (SSA/PCD). There is some ongoing controversy about what constitutes cytological dysplasia. There is consensus that cytological change resembling conventional adenomas is one form of cytological dysplasia (Fig. 27). In the past the term "mixed SSA/P – tubular adenoma" has been used for lesions with this type of dysplasia but that terminology is strongly discouraged because it ignores the molecular fact that this cytological dysplasia is occurring along a completely different molecular pathway than conventional adenomas (i.e. there is presumed to be no *APC* mutation in these lesions) and in general SSA/PCD is thought to be a more aggressive lesion than a conventional adenoma, with a high risk for the rapid development of carcinoma, even in the absence of "high grade dysplasia" from a conventional morphological perspective. For that reason (direct progression of low grade dysplasia to carcinoma), cytological dysplasia in SSA/P is not necessarily subclassified into high and low grade. The second type of dysplasia that is generally agreed upon is serrated dysplasia characterized by mitotically active, moderately pleomorphic cells that are low columnar or cuboidal and have abundant eosinophilic cytoplasm. The nuclei are generally round and open with prominent nucleoli (Fig. 28). These are clearly neoplastic appearing cells with high grade cytology. Both conventional dysplasia and this form of serrated dysplasia are commonly seen at the interface of SSA/P and carcinoma. Liu et al. have described another form of cytological dysplasia which they

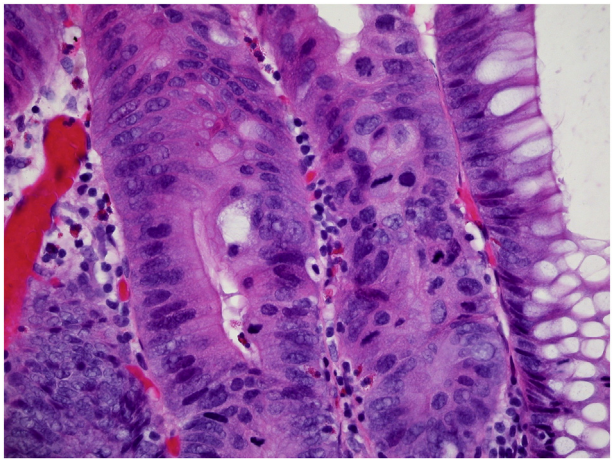

Fig. 28. Sessile serrated adenoma with serrated cytological dysplasia. The crypt on the right is background SSA. The serrated dysplasia is cytologically high grade with rounded stratified nuclei, abundant mitotic activity and eosinophilic cytoplasm.

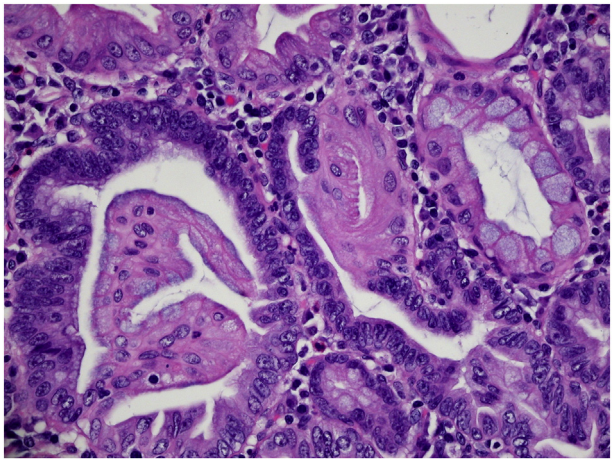

Fig. 29. Sessile serrated adenoma with unclassified cytological dysplasia according to the scheme of Liu et al. (33). The cytologically dysplasic cells differ from conventional cytological dysplasia in that the nuclei are more rounded without pseudostratification and with small nucleoli but with only minimal mitotic activity and with a small amount of cytoplasm creating a cuboidal rather than columnar cell.

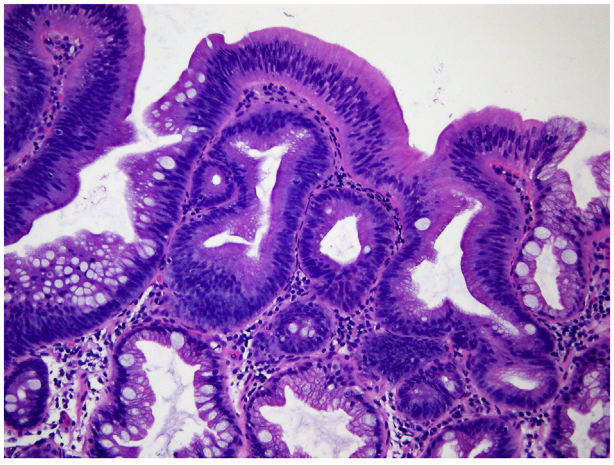

Fig. 30. Enteric metaplasia in a sessile serrated adenoma. These cells are tall cuboidal with eosinophilic cytoplasm, with oval nuclei showing some evidence of pseudostratification but show no mitotic activity. They can be seen in traditional serrated adenoma (Fig. 36), conventional tubular adenomas (Fig. 8), and SSA as well as sometimes in non-neoplastic or inflamed mucosa and in hyperplastic polyps. They are nearly identical to normal small intestinal absorptive cells (Fig. 31).

refer to as “dysplasia NOS”, which resembles conventional dysplasia but with more cuboidal cells [33] (Fig. 29). In their series this was the most common type of dysplasia. Most pathologists are likely to lump this type into conventional dysplasia. Liu et al. demonstrated a higher incidence of microsatellite instability in this group than in the other two groups. The final and not universally accepted change sometimes reported as “serrated dysplasia” is composed of tall bland cells with eosinophilic cytoplasm and oval nuclei that can appear pseudostratified but are not hyperchromatic (Fig. 30). This changes has been referred to previously as enteric metaplasia. These cells are not mitotically active and do not mark with Ki-67. These are the cells commonly seen in traditional serrated adenoma, as will be described below, and are often incorrectly referred to as dysplastic there as well. These cells were described many years ago as representing “enteric metaplasia” based on morphology and ultrastructural studies [34]. Histologically they are very similar to normal duodenal or jejunal epithelial cells and can be seen in inflamed mucosa as well as in neoplastic and hyperplastic

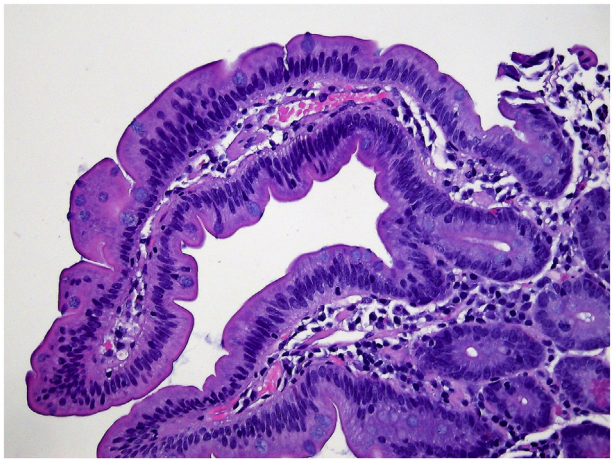

Fig. 31. Normal duodenum lined with enteric absorptive cells. Note the histological similarity to enteric metaplasia in a variety of polyp types.

polyps (Fig. 31). To the best of my knowledge no one has ever illustrated these cells converting directly to carcinoma and in my opinion they should not be categorized as dysplasia, especially in SSA where the diagnosis of cytological dysplasia implies potential for rapid progression to carcinoma. Others are in agreement with this sentiment [33]. A recent study describing these cells as dysplastic (although not specifically separating high grade serrated dysplasia as described above from what we would call “enteric metaplasia”) noted that this change was much less commonly associated with malignancy than “conventional” dysplasia, arguing against considering this change as the equivalent of more conventional appearing dysplasia [35].

3.2. Problems in diagnosis

The biggest issue in the diagnosis of SSA/P is distinguishing SSA/P from microvesicular hyperplastic polyps (MVHP) (Fig. 32). This problem occurs in large part because most SSA/P contain areas of growth that do not demonstrate abnormal crypt bases but rather have normal basal proliferation similar to that seen with MVHP. This is one of the major arguments for suggesting the possibility of SSA/P arising in MVHP. However there are also strong arguments against this possibility (including disparity in distribution of the two lesions within the colon and the occasional diagnosis of SSA/P consisting of only a few crypts

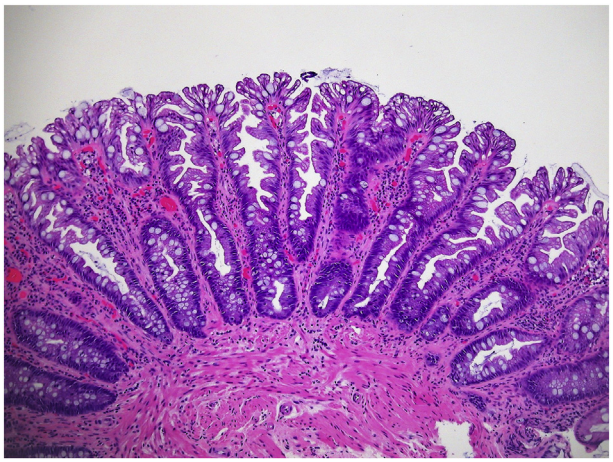

Fig. 32. Microvesicular hyperplastic polyp. At low power the MVHP has an orderly growth pattern with similar sized crypts showing proliferation at the base and increasing serration toward the lumen. Compare with the disordered overall architecture of the typical SSA (Fig. 25).

with no obvious MVHP present). At this point the origin SSA/P is not definitively known and it is possible that there are both de-novo SSA/Ps and SSA/Ps that arise from MVHP. As a rule, the presence of any truly distorted crypt should be considered enough to allow the diagnosis of SSA/P as long as that crypt is not simply a dilated crypt. The truly abnormal crypts of SSA/P should track laterally along the muscularis mucosae and should be lined with mature cells (usually either goblet cell type or gastric foveolar in appearance) without mitotic activity (Fig. 26). Perhaps too much emphasis has been placed on this definitive finding and other less well recognized findings are often ignored. Included among these are the presence of prominent serration in the deeper aspects of the crypts, but not necessarily totally basal, and marked variation in the shape and size of the crypts. HPs are usually very uniform and most serration is present near the lumen of the crypts. It should also be recognized that in the rectum there is often some crypt distortion normally in the form of elongated, occasionally branched and curved crypts that should not be mistaken for the abnormal crypts of SSA/P.

Technical issues can also cause problems. Most common among these are cauterized specimens, small biopsies of large lesions and poorly oriented specimens. If the base of the crypts is not identified in the specimen, deeper tissue levels may allow a definitive diagnosis. If the specimen is too cauterized or otherwise inadequate to allow a final diagnosis, cases can be reported as “serrated polyp, unclassified” with a comment as to why it cannot be classified. Such cases in the right colon are best managed as SSA/P and in the left colon as HPs.

3.3. Carcinoma arising in SSA/P

Carcinomas arising in SSA/P include “garden variety” adenocarcinoma (adenocarcinoma NOS), mucinous carcinoma and serrated adenocarcinoma (Figs. 33 and 34). Although carcinomas with mixed morphology and medullary carcinoma are commonly associated with the microsatellite instability pathway, these are not normally seen in the small early carcinomas detected in polypectomy specimens.

Relatively little has been written about the management of these lesions and since most SSA/P are sessile ill-defined lesion prone to incomplete resection in most cases decisions about management do not include the options available for conventional adenomas, particularly pedunculated ones. Nevertheless, it seems reasonable to use criteria similar to those for carcinomas for conventional adenoma for managing SSA/P with invasive carcinoma until additional data are available.

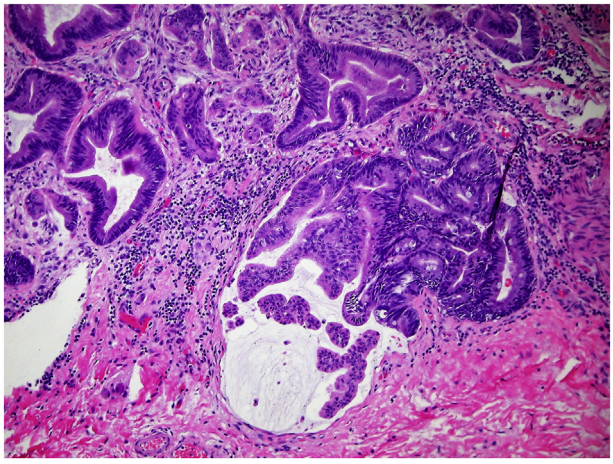

Fig. 34. SSA with early carcinoma. The tumor is already showing a serrated growth pattern with eosinophilic cells growing in a somewhat “papillary” or serrated pattern in a pool of mucin.

4. Traditional serrated adenoma

4.1. Diagnosis

Despite disagreements in the literature about the absolute diagnostic criteria for TSA, in practice there seems to be perhaps less disagreement in diagnosis than for SSA/P versus HP. The reason for this may be that for a well- developed TSA there are no real differential diagnostic possibilities except for villous adenoma, which is usually ruled out by the absence of diffuse conventional dysplasia. TSAs are typically sessile lesions in the left colon with an overall villous growth pattern (Fig. 35). Often these villi are lined with enteric metaplastic cells as described above (Fig. 36). These cells are tall columnar cells with eosinophilic cytoplasm and bland oval elongated nuclei that are sometimes pseudostratified but without hyperchromasia or mitotic activity. Sometimes a brush border is seen on these cells. TSAs with enteric epithelium typically have a very characteristic undulating border to the villi (Fig. 36), a pattern which is interestingly typical of normal small intestinal epithelium as well (Fig. 31). The presence of small abortive crypts (ectopic crypts) is a nearly ubiquitous feature (Fig. 37). We consider the presence of ectopic crypts as a defining feature of TSA [32], and a feature that can be used for diagnosis in those TSAs with a predominant goblet cell rather than enteric metaplastic appearance (Fig. 38). Although other authors have argued against ectopic crypts as

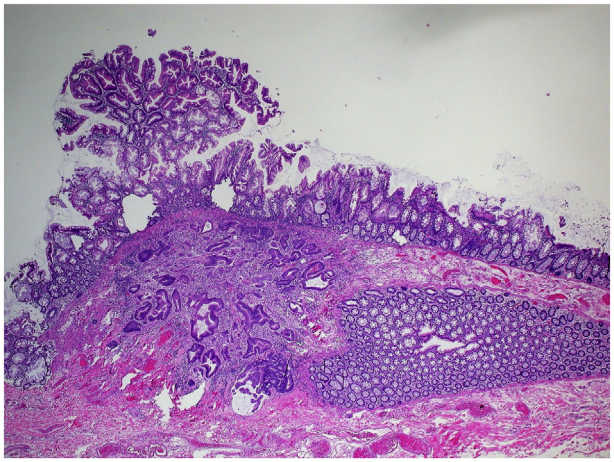

Fig. 33. SSA with early carcinoma. The tumor is arising without any evident high grade cytological dysplasia, which is typical.

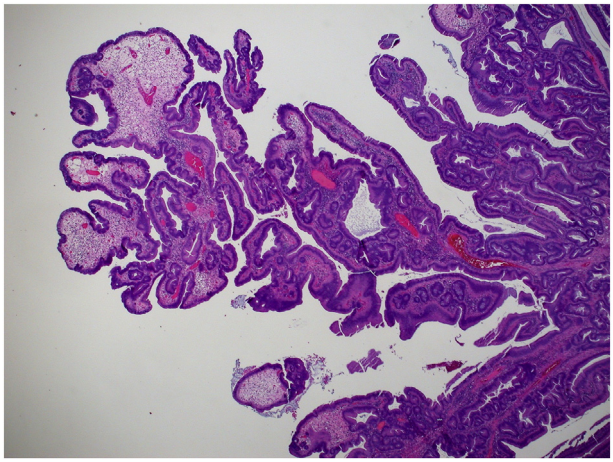

Fig. 35. Traditional serrated adenoma. This the common growth pattern with elongated villi, often with a bulbous edematous tip.

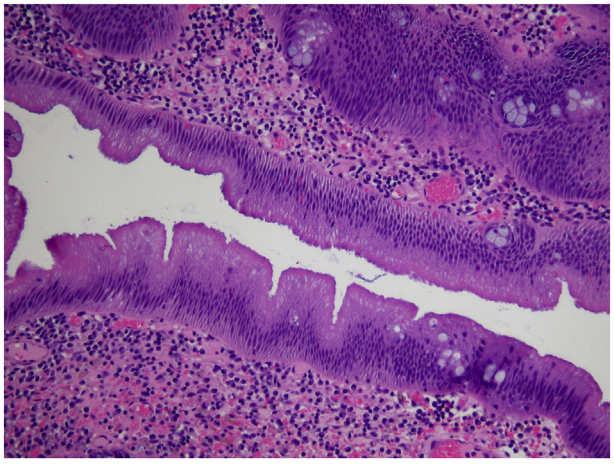

Fig. 36. Traditional serrated adenoma. Most often cells with an enteric appearance make up the bulk of the epithelium in these tumors, but sometimes goblet cells make up a large part or all of the epithelium. Note that with the enteric epithelium there is often an undulating edge to the epithelium which is very distinctive.

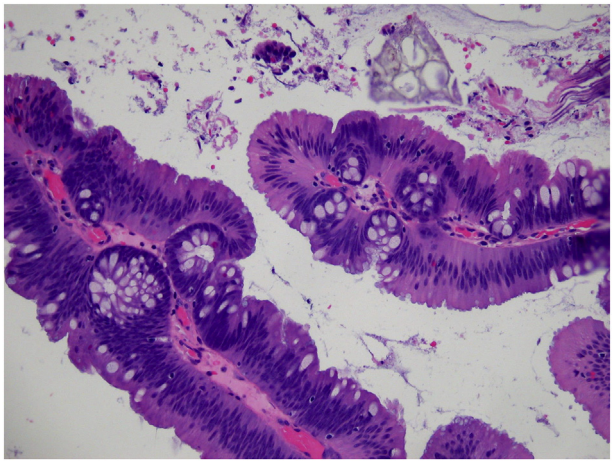

Fig. 37. Traditional serrated adenoma demonstrating numerous ectopic crypts in associated with enteric epithelium. Typically the ectopic crypts contain goblet cells even in enteric areas. Mitotic activity in TSA is generally confined to the base of these ectopic crypts and the deep aspects of the villi near the muscular mucosae.

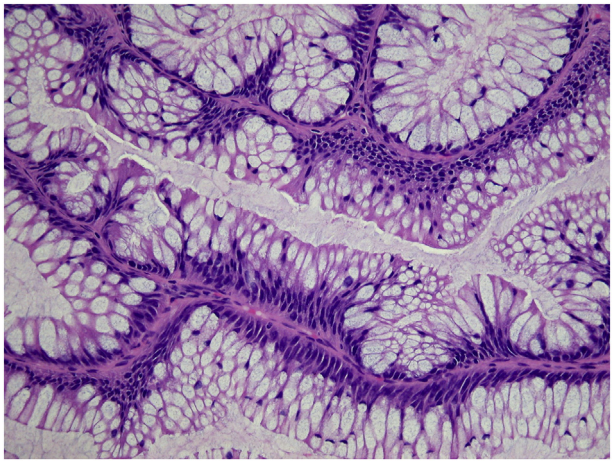

Fig. 38. Traditional serrated adenoma with preponderant goblet cells. Ectopic crypts are still present.

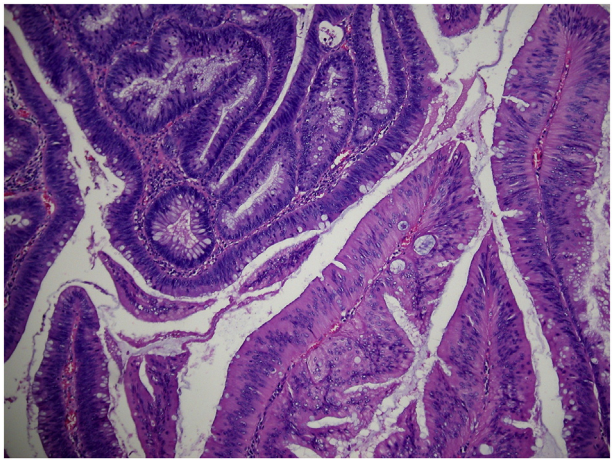

Fig. 39. Traditional serrated adenoma with conventional dysplasia. Dysplasia in TSA can be either conventional as in this case, looking like a conventional villous adenoma, or can be serrated similar to Fig. 28.

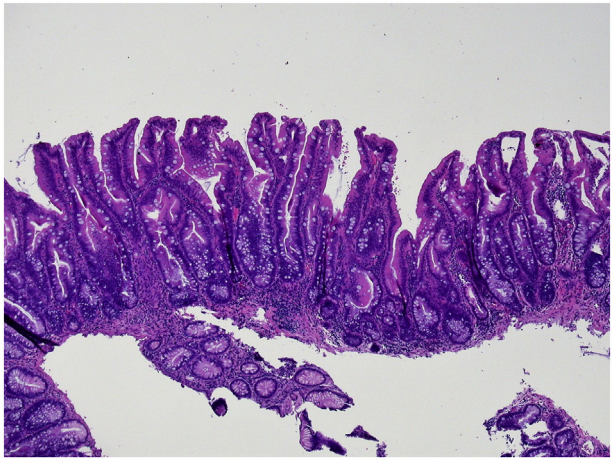

Fig. 40. Flat traditional serrated adenoma. These lesions are at first glance difficult to classify since they are composed almost entirely of enteric epithelium but with very little crypt elongation and with no prominent villi.

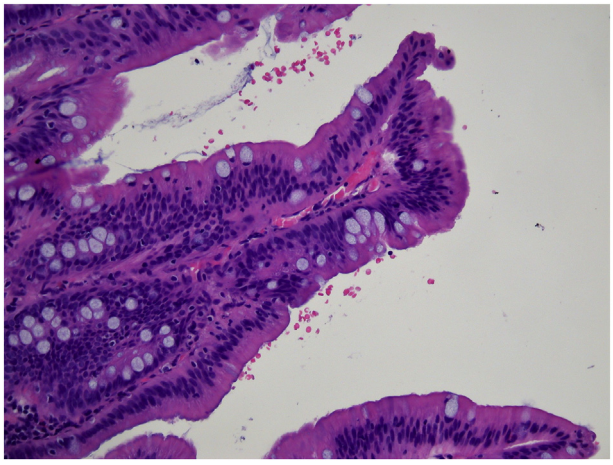

Fig. 41. Flat traditional serrated adenoma. Despite the overall flat appearance, occasional ectopic crypts can be identified.

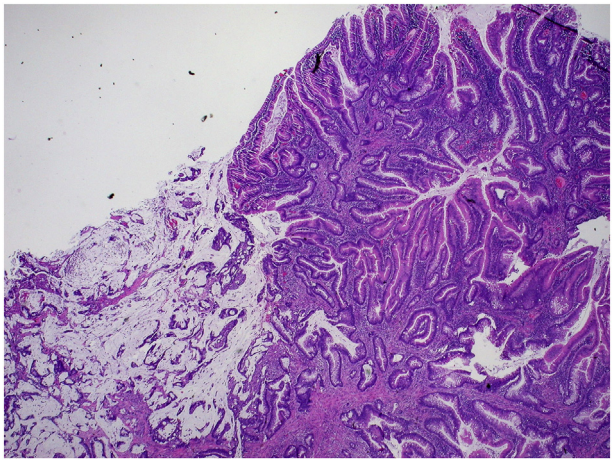

Fig. 42. Mucinous adenocarcinoma arising in traditional serrated adenoma. Carcinomas in TSA can be conventional in type (“NOS), mucinous or serrated.

a defining feature, even studies that do not “require’ ectopic crypts for diagnosis report finding them in > 90% of cases [3]. Other features sometimes seen with TSA are bulbous somewhat edematous villi sometimes described as “tennis racquet like”, and a hyperserrated appearance. Conventional cytological dysplasia with abundant mitotic activity is not found in uncomplicated TSA but does develop as a step in progression to carcinoma.

TSAs can develop cytological dysplasia usually resembling that of conventional adenomas and therefore are reported as TSA with conventional dysplasia (Fig. 39). When the conventional dysplasia is extensive the lesion takes on an appearance of a conventional villous adenoma, but careful evaluation of the entire lesion will reveal at least a few villi showing typical features of TSA. Development of conventional dysplasia seems to be a mandatory step in progression to carcinoma. The dysplasia can be low grade or high grade, and should be reported as such although at this point it is not definitively known if this distinction has clinical significance in management.

4.2. Problems in diagnosis

In general, TSA is not a difficult diagnosis. Problems usually arise in two setting: so called “flat” TSA and TSA with extensive conventional dysplasia. In addition, TSA occasionally arises apparently in an otherwise characteristic SSA/P, which may lead to confusion about management.

“Flat” TSA is characterized by very little elongation of crypts and minimal “polyp” formation. The lesion is notable as a serrated lesion with the crypts lined with enteric metaplastic cells (Fig. 40). The usual temptation is to consider the lesion an unusual hyperplastic polyp since there is little crypt distortion and the regenerative compartment appears to be normal, however close examination will reveal occasional small ectopic crypts, allowing the diagnosis (Fig. 41). These are extremely rare lesions in my experience.

TSAs with extensive conventional dysplasia are very often diagnosed as villous adenomas, which from a practical perspective may not be consequential. The only way to distinguish the two is by finding a few villi lined with enteric cell and demonstrating ectopic crypts, which is the clue to the diagnosis. Given the frequency of *KRAS* mutations in TSA, it may be that these lesions are responsible for the reported prevalence of *KRAS* in villous adenomas.

Precursor lesions for TSA have still not been well defined and one can find lesions variously categorized as goblet cell rich or microvesicular hyperplastic polyps, serrated polyps NOS or even SSA/P adjacent to TSA [3]. If the lesion is predominantly TSA then the precursor lesion is probably of little consequence. Occasionally, however, one finds a small TSA arising from an otherwise typical SSA/P. The natural history of such lesions is not well characterized but in my opinion if the TSA is a minor component and does not demonstrate conventional cytological dysplasia, these lesions should be managed as SSA/P.

4.3. Carcinoma in TSA

Carcinomas arising in TSA can be “garden variety adenocarcinoma NOS”, but can also be mucinous or serrated adenocarcinomas (Fig. 42). They are often CIMP-H but are essentially never MSI.

Carcinoma arising in TSA has not been specifically studied regarding risk factors for aggressive behavior. Therefore at this point the criteria used to evaluate carcinomas in conventional adenomas should be applied to management. Since many of these lesions are located in the rectum, localized therapy is often possible.

References

[1] Snover DC. Update on the serrated pathway to colorectal carcinoma. Hum Pathol 2011;42:1–10.
[2] Dienstmann R, Vermeulen L, Guinney J, Kopetz S, Tejpar S, Tabernero J. Consensus molecular subtypes and the evolution of precision medicine in colorectal cancer. Nat Rev Cancer 2017;17:79–92.
[3] Bettington ML, Walker NI, Rosty C, Brown IS, Clouston AD, McKeone DM, et al. A clinicopathological and molecular analysis of 200 traditional serrated adenomas. Mod Pathol 2015;28:414–27.
[4] Powell SM, Zilz N, Beazer-Barclay Y, Bryan TM, Hamilton SR, Thibodeau SN, et al. APC mutations occur early during colorectal tumorigenesis. Nature 1992;359:235–7.
[5] Pai RK, Rybicki LA, Goldblum JR, Shen B, Xiao SY, Liu X. Paneth cells in colonic adenomas: association with male sex and adenoma burden. Am J Surg Pathol 2013;37(1):98–103.
[6] Pantanowitz L. Colonic adenoma with squamous metaplasia. Int J Surg Pathol 2009;17(4):340–2.
[7] Domoto H, Terahata S, Senoh A, Sato K, Aida S, Tamai S. Clear cell change in colorectal adenomas: its incidence and histological characteristics. Histopathology 1999;34(3):250–6.
[8] Yadamsuren EA, Nagy S, Pajor L, Lacza A, Bogner B. Characteristics of advanced- and non advanced sporadic polypoid colorectal adenomas: correlation to KRAS mutations. Pathol Oncol Res 2012;18(4):1077–84.
[9] Barry EL, Baron JA, Grau MV, Wallace K, Haile RW. K-ras mutations in incident sporadic colorectal adenomas. Cancer 2006;106(5):1036–40.
[10] Fearon ER, Vogelstein B. A genetic model for colorectal tumorigenesis. Cell 1990;61(5):759–67.
[11] Ishii T, Notohara K, Umapathy A, Mallitt KA, Chikuba H, Moritani Y, et al. Tubular adenomas with minor villous changes show molecular features characteristic of tubulovillous adenomas. Am J Surg Pathol 2011;35:212–20.
[12] Neilsen JA, Lager DJ, Lewin M, Weber JJ, Roberts CA. Incidence of diagnostic change in colorectal polyp specimens after deeper sectioning at 2 different laboratories staffed by the same pathologists. Am J Clin Pathol 2013;140:213–37.
[13] Mahajan D, Downs-Kelly E, Liu X, Pai RK, Patil DT, Rybicki L, et al. Reproducibility of the villous component and high-grade dysplasia in colorectal adenomas < 1 cm: implications for endoscopic surveillance. Am J Surg Pathol 2013;37(3):427–33.
[14] Osmond A, Li-Chang H, Kirsch R, Divaris D, Falck V, Liu DF, et al. Interobserver variability in assessing dysplasia and architecture in colorectal adenomas: a multicentre Canadian study. J Clin Pathol 2014;67(9):781–6.
[15] Martinez ME, Baron JA, Lieberman DA, Schatzkin A, Lanza E, Winawer SJ, et al. A pooled analysis of advanced colorectal neoplasia diagnoses after colonoscopic polypectomy. Gastroenterology 2009;136(3):832–41.
[16] Appelman HD. Con: high-grade dysplasia and villous features should not be part of the routine diagnosis of colorectal adenomas. Am J Gastroenterol 2008;103(6):1329–31.
[17] Lieberman DA, Rex DK, Winawer SJ, Giardiello FM, Jounson DA, Levin TR. Guidelines for colonoscopy surveillance after screening and polypectomy: a consensus update by the US Multi-Society Task Force on Colorectal Cancer. Gastroenterology 2012;143(3):844–57.
[18] Mills SE, Allen Jr. MS, Cohen AR. Small-cell undifferentiated carcinoma of the colon: a clinicopathologic study of 5 cases and their association with colonic adenomas. Am J Surg Pathol 1983;7(7):643–516314828.
[19] Williams GT, Blackshaw AJ, Morson BC. Squamous carcinoma of the colorectum and its genesis. J Pathol 1979;129(3):139–47.
[20] Soga K, Konishi H, Tatsumi N, Konishi C, Nakano K, Wakabayashi N, et al. Clear cell adenocarcinoma of the colon: a case report and review of literature. World J Gastroenterol 2008;14(7):1137–40.
[21] Amin MB, Edge S, Greene F, Byrd DR, Brookland RK, Washington MK, editors. AJCC cancer staging manual. 8th ed.Springer International Publishing: American Joint Commission on Cancer; 2017.
[22] Kenney BC, Jain D. Identification of lymphatics within the colonic lamina propria in inflammation and neoplasia using the monoclonal antibody D2-40. Yale J Biol Med 2008;81(3):103–13.

[23] Tanizawa T, Seki T, Nakano M, Kamiyama R. Pseudoinvasion of the colorectal polypoid tumors: serial section study of problematic cases. Pathol Int 2003;53(9):584–90. (PubMed PMID: 14507314).
[24] Panarelli NC, Somarathna T, Samowitz WS, Kornacki S, Sanders SA, Novelli MR, et al. Diagnostic challenges caused by endoscopic biopsy of colonic polyps: a systemic evaluation of epithelial misplacement with review of problematic polyps from the bowel cancer screening program, United Kingdom. Am J Surg Pathol 2016;40:1075–82.
[25] Butte JM, Tang P, Gonen M, Shia J, Schattner M, Nash GM, et al. Rate of residual disease after complete endoscopic resection of malignancy colonic polyp. Dis Colon Rectum 2012;55:122–7.
[26] Aarons CB, Shanmugan S, Bleier JI. Management of malignant colon polyps: current status and controversies. World J Gastroenterol 2014;20:16178–83.
[27] Sonoo H, Kameyama M, Inatugi N, Nonomura A, Enomoto Y. Pedunculated polyp of early sigmoid colon cancer with invasive micropapillary carcinoma. Jpn J Clin Oncol 2009;39:523–7.
[28] Kim S, Park HK, Jung HY, Lee SY, Min KW, Kim WY, et al. ERG immunohistochemistry as an endothelial marker for assessing lymphovascular invasion. Korean J Pathol 2013;47:355–64.
[29] Haggitt RC, Glotzbach RE, Soffer EE, Wruble LD. Prognostic factors in colorectal carcinomas arising in adenomas: implications for lesions removed by endoscopic polypectomy. Gastroenterology 1985;89(2):328–36.
[30] Nascimbeni R, Burgart LJ, Nivatvongs S, Larson DR. Risk of lymph node metastasis in T1 carcinoma of the colon and rectum. Dis Colon Rectum 2002;45:200–6.
[31] Beaton C, Twine CP, Williams GL, Radcliffe AG. Systemic review and metaanalysis of histopathological factors influencing the risk of lymph node metastasis in early colorectal cancer. Colorectal Dis 2013;15:788–97.
[32] Torlakovic EE, Gomez JD, Driman DK, Parfitt JR, Wang C, Benerjee, et al. Sessile serrated adenoma (SSA) vs. traditional serrated adenoma (TSA). Am J Surg Pathol 2008;32:21–9.
[33] Liu C, Walker NI, Leggett BA, Whitehall VL, Bettington ML, Rosty C. Sessile serrated adenomas with dysplasia: morphological patterns and correlations with MLH1 immunohistochemistry. Mod Pathol 2017;30:1728–38.
[34] Yokoo H, Usman I, Wheaton S, Kampmeier PA. Colorectal polyps with extensive absorptive enterocyte differentiation: histologically distinct variant of hyperplastic polyps. Arch Pathol Lab Med 1999;123:404–10.
[35] Cenaj O, Gibson J, Odze RD. Clinicopathologic and outcome study of sessile serrated adenomas/polyps with serrated versus intestinal dysplasia. Mod Pathol 2018;31:633–42.