Deeper Examination of Negative Colorectal Biopsies

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Abstract

Initial histologic sections of specimens from colorectal biopsies of putative lesions may lack polyps. These sections may contain lymphoid aggregates that seemingly correlate with endoscopic findings; however, additional sections might contain polyps. We reviewed 83 specimens from colorectal biopsies of putative lesions for which initial sections lacked polyps. Our objectives were to determine the incidence of polyps within additional sections and to determine whether the presence of lymphoid aggregates within initial sections excludes the presence of polyps within additional sections. Eight specimens (10%) contained polyps (5 adenomatous, 3 hyperplastic), which remained histologically occult until examination to depths of approximately 120 to 380 µm. Five polyps (62%) were associated with lymphoid aggregates that were present within initial sections. We conclude that additional sections may contain surprisingly large numbers of polyps and that lymphoid aggregates present within initial sections fail to exclude the presence of polyps within additional sections.

Colorectal adenomatous polyps (adenomas) and hyperplastic polyps are extremely common. Accurate histologic interpretation of specimens from colorectal biopsies of putative lesions is important because adenomas, hyperplastic polyps, and variations of normal mucosa are endoscopically indistinguishable, and clinical follow-up is determined in part by identification and classification of colorectal polyps. Initial histologic sections of specimens from biopsies of putative lesions may lack polyps. These sections may contain lymphoid aggregates that seemingly correlate with endoscopic findings; however, additional sections might contain polyps. The incidence of polyps that are revealed by examining additional sections is unknown.

We studied specimens from colorectal biopsies of putative lesions for which initial sections lacked polyps to determine the incidence of polyps within additional sections and to determine whether the presence of lymphoid aggregates within initial sections excludes the presence of polyps within additional sections.

Materials and Methods

We prospectively studied 90 consecutive formalin-fixed, paraffin-embedded specimens from colorectal biopsies of putative lesions for which initial sections lacked polyps during the interval January 1, 2001, through May 31, 2001. We then examined 50 additional sections for every specimen included in the study. Initial sections were obtained from a continuous ribbon of 20 well-oriented, 4-µm-thick, H&E-stained consecutive serial sections. These sections were placed on 2 slides, 10 sections per slide. Initial sections...
enabled examination of specimens to depths of approximately 80 µm. Additional sections were obtained from a continuous ribbon of 75 well-oriented, 4-µm-thick, H&E-stained consecutive serial sections. From this ribbon, 50 sections were retained, and 25 sections were discarded, by repeating the process of placing 10 consecutive serial sections on a slide and discarding the next 5 consecutive serial sections. The combination of initial and additional sections enabled examination of specimens to depths of approximately 380 µm. All biopsies were performed or supervised by experienced endoscopists at the University of California Los Angeles School of Medicine, were performed with jumbo forceps, and were performed without cautery. We each have special interest in gastrointestinal pathology, and we all reviewed all slides.

The histologic presence or absence of lymphoid aggregates and polyps was noted for each specimen. Lymphoid aggregates were defined as clusters, nodules, or follicles of lymphocytes that were easily appreciated when magnified a total of ×40. Polyps were defined in part according to standard criteria. Adenomas were defined as well-circumscribed foci of dysplastic epithelium; and hyperplastic polyps were defined as well-circumscribed foci of markedly serrated crypts with nondysplastic epithelium, associated or unassociated with thick subepithelial collagen. For our study, only dysplasia or serration easily appreciated by all of us when magnified a total of ×40 was considered definitive. If we agreed that additional sections displayed changes indeterminate for adenomas or hyperplastic polyps, a second set of 75 additional sections was obtained in the same manner that was used for the first set of additional sections, to examine these specimens to depths of approximately 680 µm. If changes remained indeterminate, the specimens were considered negative for polyps.

For all 90 specimens, if diagnostic disagreement occurred among us, the specimens were excluded from further investigation.

### Results

The 90 specimens were from 75 patients, 16 patients having multiple specimens. Seven specimens (8%) were excluded owing to diagnostic disagreement among us. Diagnostic disagreement involved regeneration vs dysplasia, hyperplasia vs hyperplastic polyp, mucosal prolapse vs normal, and leiomyoma vs normal. Three specimens displayed changes such as minimal nuclear pseudostratification or minimally serrated crypts within the first set of additional sections that we all agreed were indeterminate for adenomas or hyperplastic polyps, and for these specimens further additional sections were obtained.

#### Table 1

<table>
<thead>
<tr>
<th>Polyp</th>
<th>Lymphoid Aggregates on Initial Slides?</th>
<th>Serial Sections Required</th>
<th>Depth of Sectioning Required (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hyperplastic</td>
<td>No</td>
<td>60-65</td>
<td>240-260</td>
</tr>
<tr>
<td>Hyperplastic</td>
<td>Yes</td>
<td>60-65</td>
<td>240-260</td>
</tr>
<tr>
<td>Adenomatous</td>
<td>No</td>
<td>60-65</td>
<td>240-260</td>
</tr>
<tr>
<td>Adenomatous</td>
<td>Yes</td>
<td>90-95</td>
<td>360-380</td>
</tr>
<tr>
<td>Hyperplastic</td>
<td>Yes</td>
<td>30-35</td>
<td>120-140</td>
</tr>
<tr>
<td>Adenomatous</td>
<td>Yes</td>
<td>45-50</td>
<td>180-200</td>
</tr>
<tr>
<td>Adenomatous</td>
<td>Yes</td>
<td>60-65</td>
<td>240-260</td>
</tr>
<tr>
<td>Adenomatous</td>
<td>No</td>
<td>60-65</td>
<td>240-260</td>
</tr>
</tbody>
</table>

For the entire study, additional sections for 8 specimens (10%) contained definitive histologic features of polyps. These polyps were from 8 patients, 1 polyp per specimen per patient. Five polyps were pure tubular adenomas, and 3 were pure hyperplastic polyps. The polyps remained histologically occult until examination to depths of approximately 120 to 140 µm (1 hyperplastic polyp), 180 to 200 µm (1 adenoma), 240 to 260 µm (3 adenomas, 2 hyperplastic polyps), or 360 to 380 µm (1 adenoma). Of the 8 patients who had polyps detected, 3 (38%) were without previously or concurrently diagnosed polyps or adenocarcinoma, and 5 (62%) had previously or concurrently diagnosed polyps or adenocarcinoma.

Five (62%) of 8 polyps were associated with lymphoid aggregates that were present within initial sections Table 1, Image 1, and Image 2.

Other focal findings that frequently were present within specimens that lacked polyps included active cryptitis, excessive chronic inflammation, edema, hemorrhage, excessive collagen, elongated crypts, architectural disarray, innominate glands, fecal botanical elements, and fibroadipose tissue. Occasionally these findings were absent from initial sections and were revealed on examination of additional sections. Some of these focal findings also were associated with the polyps (Image 2). If features of adenomas or hyperplastic polyps were absent, we refrained from sectioning beyond the first set of additional sections. Presumably, some of these changes accounted for endoscopic findings. However, we often disagreed as to which histologic findings adequately explained endoscopic findings or as to which histologic findings were variations of normal.

### Discussion

Examination of initial histologic sections can fail to confirm clinically suspected colorectal polyps. Initial histologic sections can contain lymphoid aggregates that mimic small or diminutive polyps endoscopically. We studied 83 specimens...
from colorectal biopsies of putative lesions for which initial sections lacked polyps. Examination of additional sections revealed polyps for nearly 10% of specimens. Furthermore, exhaustive sectioning might have revealed more polyps. Similar results were described by Calhoun et al,5 who detected polyps in a high percentage of cases on examining additional sections obtained from the reverse sides of the specimens.

Indications for additional sections are best viewed from the perspectives of clinicopathologic correlation, financial cost, and clinical significance. The broad differential diagnosis of small or diminutive putative colorectal lesions includes epithelial proliferations, mesenchymal proliferations, inflammatory processes, artifacts, fecal elements, and variations of normal mucosa.4 Because these diagnostic considerations may be endoscopically indistinguishable, accurate diagnosis requires histologic confirmation. Therefore, if initial sections lack polyps, examination of additional sections improves the accuracy of diagnosis and the accuracy of clinicopathologic correlation.

In our study, many patients who had polyps detected had previously or concurrently diagnosed polyps or adenocarcinoma, and the finding of an additional polyp likely would have little influence on treatment or prognosis. It might be cost-effective to examine additional sections only when positive results would significantly influence treatment or prognosis. Polyps distal to the splenic flexure, adenomatous or hyperplastic, of any size found at flexible sigmoidoscopy may portend increased risk of advanced proximal neoplasia and may prompt pancolonoscopic screening and frequent surveillance.2,6,7 Therefore, if initial sections lack polyps, additional sections are indicated for patients without previously or concurrently diagnosed polyps or adenocarcinoma.

The optimal depth of sectioning required to adequately investigate initially negative specimens remains to be fully

Image 1  A, Initial section from putative lesion contains normal mucosa with lymphoid aggregate. B and C, Additional sections contain hyperplastic polyp and lymphoid aggregate. (H&E, original magnification ×40)
determined. Theoretically, thorough examination requires exhaustive sectioning. However, benefits of exhaustive sectioning must be weighed against direct and indirect costs. In our study, 7 (88%) of 8 polyps were detected with examination to depths of 240 to 260 µm, and all 8 polyps were detected with examination to depths of 360 to 380 µm. These results suggest that examination to depths of 240 to 260 µm detects the majority of histologically occult polyps and is potentially cost-effective. By following this guideline, only approximately 10% of polyps might remain histologically occult.

Of note, the financial cost of obtaining additional sections compares favorably with that of other commonly ordered ancillary studies, such as immunostains.

In our study, most polyps that were revealed within additional sections were associated with lymphoid aggregates that were present within initial sections. Consequently, the presence of lymphoid aggregates within initial sections fails to exclude the presence of polyps within additional sections. Therefore, if polyps are suspected endoscopically, additional sections should be considered if initial sections lack polyps and contain lymphoid aggregates.

Conclusions

Initial sections of specimens from colorectal biopsies of putative lesions may lack polyps and may contain lymphoid aggregates. Additional sections may contain surprisingly large numbers of polyps. The presence of lymphoid aggregates within initial sections fails to exclude the presence of polyps within additional sections. If initial sections lack polyps, examination of additional sections improves the accuracy of diagnosis and the accuracy of clinicopathologic correlation.
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References


