Mr D J Oakland
(Queen Elizabeth Hospital, Birmingham)

The Diagnosis of Carcinoma of the Colon by Exfoliative Cytology

In practice the diagnosis of colonic disease usually depends upon the radiological appearance of the colon after barium enema or similar techniques. In most cases the rectum and possibly the distal sigmoid can be inspected directly by sigmoidoscopy and a biopsy taken of any mucosal lesion but, not infrequently, the diagnosis of cancer can neither be excluded nor proved by these methods.

Examination of material from the large bowel for the presence of exfoliated malignant cells can establish the presence of a carcinoma where other investigations are equivocal or negative. Despite the wide application of exfoliative cytology to the problem of cancer diagnosis in the last two decades (von Haam 1962), there have been few reports on the use of this technique in the colon. Those results which have been published confirm the value of the method (Raskin *et al.* 1959, Oakland 1961, Burn 1961) but all emphasize that the main difficulty of using this technique is in cleansing the bowel of its fæcal content prior to the collection of specimens. This problem is still not solved and is probably the reason why the method does not find advocates where cancer cytodiagnosis of other organs is established.

Preparation of the Patient

Early work showed conclusively that, if reliable results were to be obtained, the colon must be cleansed of its fæcal debris. This is difficult in cases with suspect lesions of the right colon but most of the diagnostic problems in which cytology is helpful are situated in the sigmoid or rectosigmoid and it is relatively easy to cleanse the distal half of the colon completely.

The following preparation, starting two days prior to examination is usually adequate although it may have to be continued for longer:

1st day : Saline enemas night and morning. Bisacodyl tablets 1 t.d.s.
2nd day : Saline enemas night and morning. Bisacodyl tablets 1 t.d.s.
3rd day : Saline enema in early morning and then collection of cytological specimen.

Collection of Specimens

Specimens are usually collected by colonic lavage with normal saline (Oakland 1961), but in a few cases a modification of the rectal washing apparatus, originally described by Loeb & Scapier (1951) is used during sigmoidoscopy.

Interpretation of Smears

Smears are reported as negative, positive or unsatisfactory. This simple classification, although inadequate from the cytological point of view, is of most value to the clinician.

Results of Colonic Exfoliative Cytology (Table 1)

Over the four years 1959–62 colonic exfoliative cytodiagnosis has been available to the clinicians of the United Birmingham Hospitals when the diagnosis was in doubt after radiological and sigmoidoscopic examination. Ninety-seven patients were examined and 24 were subsequently proved to have a carcinoma, 22 of which were situated in the left side of the colon. In 18 of these, malignant cells were found in material obtained from the colon and the diagnosis was first established by colonic cytology. Seventy-three had negative cytology and the absence of a common lesion established by laparotomy and a follow-up for a minimum of twelve months.

Table 1
Results of colonic cytology (1959–62)

Total number of patients examined	97
Total number of examinations	99
Final diagnosis not established	2
Patients with cancer	*24*
Cytology positive	18
Cytology negative (false negative)	6
Patients who did not have cancer	*73*
Cytology negative	73
Cytology positive (false positive)	0

Indications for Colonic Exfoliative Cytology

In my experience colonic cytology is of most value in the detection of overt malignant disease of the sigmoid and rectosigmoid. The majority (92%) in my series were problems referable to the distal colon where radiological and sigmoidoscopic examination had been inconclusive. Most carcinomas of the large bowel arise in the sigmoid and rectosigmoid and yet this area may contain a 'blind spot' which is difficult to visualize by radiology and it may be impossible to manipulate a sigmoidoscope past the rectosigmoid angle to obtain a direct view and a biopsy specimen.

When only a partial view of a suspicious lesion, beyond the end of the sigmoidoscope, can be obtained so long as the bowel is kept fully inflated, but is lost when the glass is removed to pass a biopsy instrument, then exfoliative cytology gives good diagnostic material.

Case 1
E R, woman, aged 60
Four months' history of bleeding per rectum and occasional diarrhœa. Examination per rectum: ? ulcer on posterior wall of rectum. Sigmoidoscopy to 15 cm showed no abnormality, apart from a small internal hæmorrhoid. Barium enema was normal. Cytology showed numerous malignant cells and at laparotomy a small polypoid adenocarcinoma was found in the sigmoid colon.

The differential diagnosis of carcinoma and diverticulitis is notoriously difficult even at operation and occasionally the two conditions co-exist. The presence of malignant cells in material washed from the suspect area of the bowel makes the diagnosis.

Case 2
W D, woman aged 77
Three months' history of left-sided abdominal pain and back pain. She also complained of melæna. Examination revealed no abnormality in the abdomen but on rectal palpation an indefinite mass could be felt about 10 cm from the anus. A barium enema showed diverticulosis of the sigmoid and a narrowed segment in the apex of the sigmoid loop (Fig 1). Sigmoidoscopy in the out-patient clinic revealed a normal bowel up to 10 cm but there was some dark blood in the lumen. Repeat sigmoidoscopy showed a small polypoid lesion in the sigmoid colon together with œdematous mucosa which appeared to be part of an inflammatory lesion. Three biopsies taken from this area were reported as showing small adenomatous polypi of the colon without evidence of malignancy. A repeat barium enema showed three sigmoid diverticula containing barium residues from a previous enema (Fig 2), together with a narrow area without any hold-up in the flow of the barium which was thought to be due to diverticulitis. Specimens were collected for exfoliative cytology and all these contained numerous adenocarcinoma cells (Fig 3). Laparotomy was performed and the patient was found to have a cicatrizing growth in the lower sigmoid. A left partial colectomy was performed.

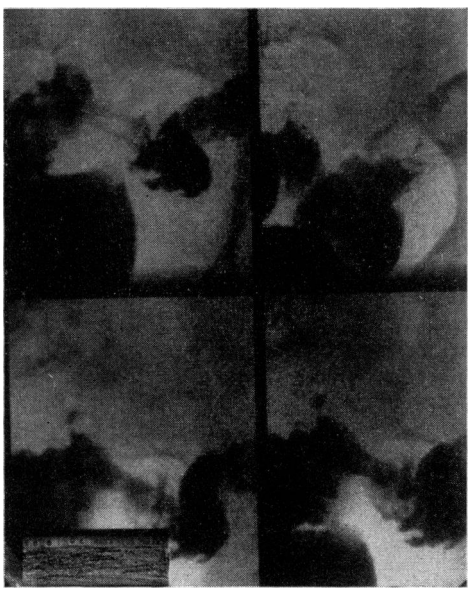

Fig 1 **Case 2** *Barium enema. Narrowed segments and diverticulosis of the sigmoid*

Fig 2 **Case 2** *Barium enema. Barium residues in sigmoid diverticula*

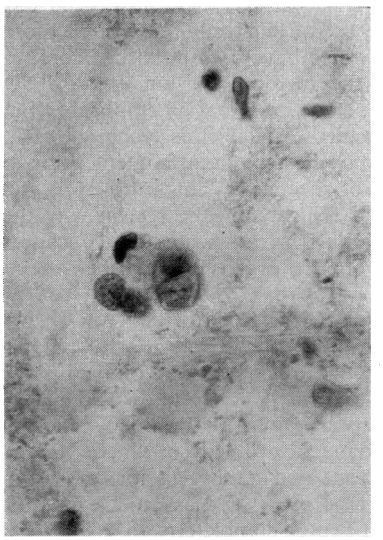

Fig 3 **Case 2** *Malignant cells in smear*

Polypi present a difficult diagnostic problem not only because biopsy, unless it is total, may not include an area of malignant degeneration but also because symptoms may be attributed to a polyp which in fact arises from an overt lesion high up, as in Case 3.

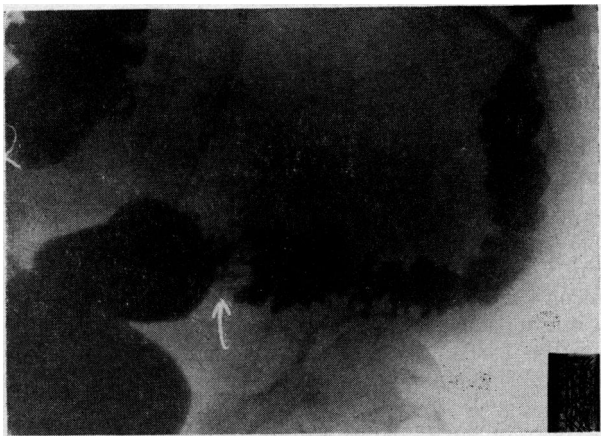

Fig 4 **Case 3** *Barium enema. Sigmoid diverticulitis*

Case 3
W B, man aged 64
One month's bleeding per rectum. Nothing abnormal on examination. Sigmoidoscopy to 15 cm showed no abnormality. Barium enema showed sigmoid diverticulitis but neoplasm could not be excluded (Fig 4). A second sigmoidoscopy showed a polypoid lesion at 15 cm. A biopsy was reported as an adenomatous polyp of large bowel type and after further sections had been cut overt carcinoma was not identified.

Cytology was strongly positive (Fig 5). At laparotomy the lower sigmoid and rectosigmoid were thick walled and it was difficult to palpate the polypoid lesion though a mass could be felt within the bowel about 8 cm from the pelvic floor. This area was resected and when the specimen was opened a carcinomatous ulcer was found as well as the more distal polyp. The bowel itself was the site of diverticulitis.

My series includes a few in whom an emergency colostomy has been established for acute and chronic obstruction of the colon. The nature of the obstructing lesion was not established at the time of the emergency operation because of dense adhesions and in one case a pelvic abscess was entered. Subsequent radiological examination of the isolated distal bowel may not help to differentiate a malignant and inflammatory lesion or they may co-exist. Cytology has helped to establish the diagnosis, as in Case 4.

Case 4
Man, aged 58
An emergency transverse colostomy was performed for acute upon chronic intestinal obstruction in December 1959. The nature of the obstructing lesion was not ascertained at operation owing to dense

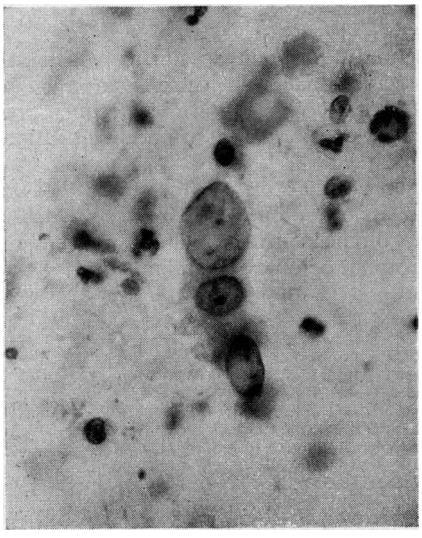

Fig 5 **Case 3** *Malignant cells in smear*

Fig 6 **Case 4** *Malignant cells in smear*

adhesions. He subsequently developed a pelvic abscess which was drained. Investigations by X-ray, sigmoidoscopy and biopsy up to June 1960 showed an obstruction at the rectosigmoid region which was thought to be inflammatory. Cytology in July 1960 was positive for malignant cells (Fig 6). Subsequent laparotomy and resection of sigmoid colon revealed a small carcinoma and extensive diverticulitis.

There seems no doubt that chronic ulcerative colitis is pre-cancerous and exfoliative cytology could be helpful in establishing the presence of a concomitant malignant lesion. It is more difficult to cleanse the bowel in these cases and as a rule total colonic lavage is required since radiological evidence of the possible site of a neoplasm is not usually available. The smears obtained from the patient contain large numbers of cells and careful screening is required.

Case 5

M R, woman, aged 45

Two months' history of intermittent abdominal pain and diarrhœa. She had passed blood per rectum just before admission. Barium enema showed an obstruction in the sigmoid colon and what appeared to be colitis of the colon proximal to this as far as the mid transverse colon (Fig 7). Sigmoidoscopy to 17 cm was not helpful. The cytology of colon material was extremely cellular, but a few groups of malignant cells were seen. At operation the appearance of the colon was more in keeping with an inflammatory lesion. The transverse descending and pelvic colon was excised. Examination of the bowel showed extensive chronic ulcerative colitis and what appeared to be an inflammatory stricture of the sigmoid. Further examination of the stricture, however, showed that it was the site of a differentiated adenocarcinoma.

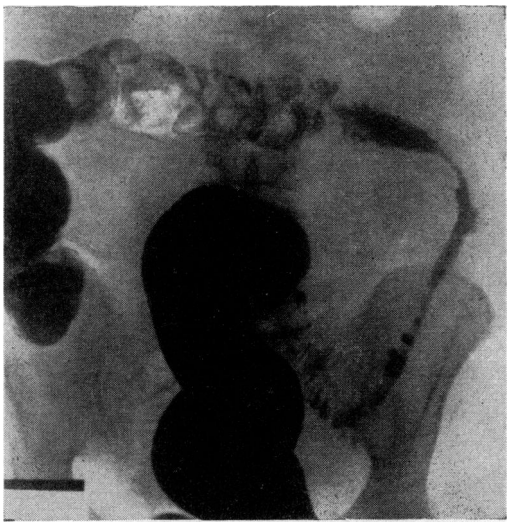

Fig 7 **Case 5** *Barium enema. Obstruction at sigmoid and colitis of proximal colon*

Other Methods of Processing and Staining Cytological Material

The difficulty of cleansing the colon of its fæcal content has encouraged cytologists to look for methods of separating the cellular from the acellular content of fæces.

Sieve and millipore filtration: Cameron & Thabet (1959) and Burn (1961) have used a membrane filter technique to separate the cellular content of colon washings. The specimen is pre-filtered through a stack of graded mesh sieves prior to passing the material through the millipore filter. I have used this technique and there is no doubt that good quality cell specimens are obtained and since they are all on one paper this reduces the time required to scan the material. However, this technique in no way reduces the amount of preparation required since the fine particulate content of fæces will pass through all the sieves and be deposited on the pores of the millipore membrane.

Acridine orange staining and ultraviolet microscopy: This staining technique is recommended by von Bertalanffy *et al.* (1956) and is quicker than the Papanicolaou technique (1942) and satisfactory fluorescence is obtained in material from the colon (Oakland 1964). However, the hope that the scanning time for each slide would be greatly reduced was not realized, since all the cellular content of the smear and a good deal of the noncellular took up the acridine orange and fluoresced under ultraviolet light. Although the malignant cells fluoresced brightly, the contrast between these cells and nonmalignant cells was not sufficient to allow for rapid scanning.

Summary

It is essential to obtain specimens which are colourless and opalescent since even a slight degree of brown coloration in the specimen produces a large deposit of fæces after centrifugation.

Unlike that of the stomach, the cytology of the colon is relatively simple, since any cell which is found in colonic washings will have been exfoliated from the colonic or rectal mucosa.

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