USES OF WHITE CELL SCINTIGRAPHY IN IBD

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Inflammatory bowel disease (IBD) is a common diagnosis which is responsible for a considerable proportion of the workload of many gastroenterologists and some surgeons. The speciality of Nuclear Medicine can provide useful clinical information in many aspects of the care of patients suffering from this condition, largely through the use of radio-isotope labelled white cell scans. These scans are not widely available, but are becoming increasingly recognised as being a valuable screening tool in the assessment of the extent and activity of IBD, and in some situations, the detection of its complications. This article defines the development of the technique, the practicalities involved and attempts to place radio-labelled white cell scanning in its current clinical context.

HISTORICAL BACKGROUND

The identification of foci of inflammation and/or infection in patients has been a persistent challenge to the clinician and before the development of radiology, clinicians relied on the patient's symptoms and physical signs as diagnostic aids. In the second half of this century, serological and haematological parameters become available to assess disease activity. The introduction of imaging modalities such as ultrasound and, in particular, CT X-ray and MRI have revolutionised patient care over the last two decades. Despite the increasingly impressive anatomical detail offered by these studies, the localisation of areas of infection and inflammation in the body continues to be a common and difficult problem to solve.

In theory, this situation should provide considerable potential for Nuclear Medicine to exploit as a predominantly functional imaging modality, as opposed to the anatomical imaging provided by most forms of conventional radiology. As more became known about the cellular and immunological response in inflammation, considerable interest was exposed in the concept of attaching a radio-isotope label to white blood cells. As a consequence, the localisation of these labelled white cells at the sites of disease or tissue damage could, in theory, be detected. This simple statement in fact conceals a number of very important practical hurdles and theoretical problems which, to some extent, remain today and which have to be overcome. Firstly, it must be possible to obtain a population of white blood cells from the patient which can then be labelled. Next, a radiopharmaceutical is required with which to label the white cells - a radiopharmaceutical is a chemical compound which effectively acts as a carrier and to which a radioactive isotope can be attached. This radiopharmaceutical should have well-documented physical and chemical properties, be avidly and perhaps selectively taken up by the white cells, and should remain attached to the white cells for the duration of the period of the study. The presence of the radiopharmaceutical on or in the white cells should not alter their usual behaviour and functional activity. Finally, the isotope used should be easy to image with currently-available gamma camera technology and should be associated with an acceptably low radiation dose to the patients undergoing the study.

The first published studies in this field were by McAfee and Thakur in 1976,[1] using predominantly the radiopharmaceuticals 111-Indium oxine and 111-Indium tropolonate. It became apparent that the major attractions of white cell scanning, in addition to its sensitivity and specificity, were that it was a relatively non-invasive procedure and could therefore be applied to all groups of patients from ambulant out-patients to those in ITU, and it also afforded the opportunity of whole body imaging which few, if any of the conventional radiological imaging modalities could offer.

In the context of IBD, labelled white cell scans are of considerable value in several clinical situations. Labelled white cell scans should not be regarded as diagnostic tests specific to IBD, as any form of intestinal inflammation or infection will result in accumulation of white cells and, in theory, a positive scan. In the early studies of labelled white cells using 111-Indium, the results showed a close correlation with radiology,[2] endoscopy and histology[3] for disease extent and activity in patients with IBD. The technique was also shown to be of value in detecting intra-abdominal[4] and intra-hepatic[5] abscesses associated with Crohn's disease. Its accuracy was confirmed when it was used as a screening test for IBD in patients with abdominal pain and/or diarrhoea.[6] The sensitivity and specificity of the technique were further improved by refinement of the cell separation and labelling process which permitted labelling of a relatively pure population of granulocytes rather than the mixed population of platelets, red cells, lymphocytes and granulocytes which had been labelled previously.[7]

Another avenue of interest with regard to 111-Indium labelled white cell scanning has been its use as a means of quantifying disease activity. The first studies involved faecal collections from patients over a number of days following injection of 111-Indium labelled white cells. The activity of 111-Indium in the faecal collection was then expressed as a percentage of the administered dose.[8,9] As might be expected, this technique was beset by technical problems surrounding the stool collection; it was also time-consuming, and generally proved to be unpopular with staff and patients alike. A variation of this technique which did not necessitate a stool collection was the estimation of whole body retention of 111-Indium labelled white cells using an uncollimated gamma camera or a whole body monitor.[10]

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These techniques are undoubtedly accurate, but they require considerable time input from senior medical personnel and as a consequence have fallen out of favour with clinicians, who have increasingly turned to laboratory markers of inflammation (such as the ESR and CRP) as surrogate markers of disease activity.

Whilst 111-Indium labelled white cell scintigraphy became an established technique in the investigation of subjects with suspected or proven IBD, in the early 1980s a number of laboratories sought alternative methods which did not necessitate the use of 111-Indium as the radio-isotope label. The main drawbacks of 111-Indium are its limited availability, poor image quality, high cost and the considerably high radiation dose to the patients as a consequence of its long half-life. The most attractive alternative to 111-Indium is 99m-Technetium. This isotope has become the mainstay of nuclear medicine imaging over the last two decades: it is relatively inexpensive and widely available, usually in the form of the decay product of a 99-Molybdenum generator. 99m-Tc has a high photon flux, and the photon energy of 140keV is ideally suited to the operating characteristics of most modern gamma cameras. In addition, the relatively short half-life of six hours results in a low radiation dose to the patient.

Early attempts using 99m-Tc as a label were fraught with problems. Wistow *et al.*[11] who attempted to complex 99m-Tc with oxine encountered problems with poor chemical stability. Others attempted borrowed from the established technique for labelling red blood cells and used reducing agents such as stannous pyrophosphate prior to the addition of 99m-Tc.[12,13] These *in vitro* studies revealed unacceptably low labelling efficiencies and were not pursued clinically. A further approach first reported by Scroth *et al.* in 1981[14] and subsequently by Danpure *et al.*[15] was the incubation of 99m-Tc labelled colloid molecules with white cells, the theory being that the white cells would phagocytose these macromolecules. Initial results of *in vitro* studies using this technique were encouraging, but it became apparent that *in-vivo* studies were likely to be handicapped by a relatively low labelling efficiency and, in particular, white cell activation resulting in significant lung margination of the white cells and, therefore, low target-to-background ratios.[16]

A major breakthrough was made by Peters *et al.*[17] who reported the first successful clinical studies with the radiopharmaceutical 99m-Tc hexamethylpropylene-amineoxime (HMPAO) as a label for white cells. HMPAO is a lipophilic compound which had been developed by Amersham International, originally for brain imaging.[18,19] Peters *et al.* attempted to utilise these lipophilic properties to permit the efficient labelling of white blood cells without causing the cell to become activated, a limitation which had plagued the other methods outlined above.

After this initial breakthrough, a number of groups studied the technique of 99m-Tc HMPAO white cell scintigraphy in a variety of clinical settings. In the context of IBD, much of the early published data came from Peters *et al.*[17] who published the first comparative studies between 99m-Tc HMPAO and 111-Indium labelled white cells, with particular emphasis on differences in labelling efficiency, biodistribution and kinetics.[20] They found that 99m-Tc HMPAO is an efficient leucocyte label, and labels granulocytes with greater stability than it does with mononuclear leucocytes. They concluded that with respect to granulocyte kinetics and clinical data, 99m-Tc HMPAO labelled leucocytes are comparable with 111-In-tropolonate labelled granulocytes; at this stage, 111-Indium labelled white cell scintigraphy was regarded as the 'gold standard' against which the new technique had to be validated.

Li *et al.*[21] performed white cell scintigraphy in 234 patients, 86 of whom were suspected to have IBD and 148 in whom the diagnosis of IBD was already established. They found that the sensitivity, specificity and accuracy of the 99m-Tc leucocyte scan were 96, 97 and 97% respectively, in comparison to 96, 97 and 97% respectively for 111-Indium leucocyte scans. It should however be noted that of the 234 patients, 146 had 99m-Tc HMPAO scans, 82 had 111-Indium labelled scans, whilst only six had both. A more direct comparison of the two techniques in the same patient was provided by Allan *et al.*[22] In this study 47 patients, 29 with active IBD on clinical and laboratory grounds, and 18 with presumed irritable bowel syndrome underwent simultaneous 99m-Tc HMPAO and 111-In oxine labelled white cell scans. All 27 patients with active disease had positive scans with both agents and there were no false positive studies. Both agents revealed similar patterns of intensity and extent of disease activity. These findings, in conjunction with the more favourable isotope availability, radiation dosimetry and image quality led the authors to conclude that 99m-Tc HMPAO is the agent of choice in detecting active IBD and that a negative scan reliably excludes active disease. The one criticism of this study is that it did not have access to radiological or endoscopic findings in the patients to allow more complete evaluation of their disease extent.

Other groups have looked at the correlation between 99m-Tc HMPAO white cell scintigraphy and radiological imaging studies. Kennan and Hayward[23] in a retrospective study looked at 18 patients with known small bowel Crohn's disease who underwent 99m-Tc HMPAO scintigraphy and had small bowel barium studies performed within the preceding nine months. They found excellent correlation between the two techniques with only one false positive and no false negative studies. These positive findings were confirmed by Scholmerich *et al.*[24] and also included patients with inflammatory large bowel disease. This study also found excellent correlation between extent of disease at colonoscopy and white cell scintigraphy, although the sample number was small.

Conflicting data as to the clinical usefullness of white cell scintigraphy was published by Gibson *et al.*[25] who looked into the value of positive scans using either 99m-Tc stannous colloid or 99m-Tc HMPAO as the radiopharmaceutical. Compared with clinical, laboratory, radiological and endoscopic techniques, they found that patients without IBD and with a low suspicion of inflammation had a high false positive rate. The results were found to be independent of the 99m-Tc cell labelling technique used. Patients with known IBD also had significant false positives with the 99m-Tc. This study also commented on the early appearance of 99m-Tc HMPAO in the bowel, in many cases within the first hour after re-injection of the labelled leucocytes.

These somewhat controversial findings did not signal the end of 99m-Tc HMPAO white cell scintigraphy, the poor results were attributed to some extent to the use of 99m-Tc stannous colloid as the radiopharmaceutical in the majority of the patients in this study. Other groups had

reported that the physiological excretion of 99m-Tc HMPAO to be a potential problem in the interpretation of labelled white cell scans using this agent,[26,27] and the unacceptably high false positive rates reported by Gibson *et al*. resulted in greater attention being paid to the imaging protocol to reduce this effect. Neilly and Gray[28] suggested a practical step to reduce the enterohepatic excretion of 99m-Tc HMPAO by fasting the patients undergoing white cell scintigraphy. This would in principle allow pooling of the 99m-Tc HMPAO in the gall bladder. In non-fasted patients, the presence of food in the stomach provokes the secretion of CCK which, in turn, causes gall bladder contraction with consequent discharge of accumulated 99m-Tc HMPAO into the small bowel. Neilly and Gray[28] were able to demonstrate that fasting resulted in a much higher number of studies in which the gall bladder was visualised, although they commented that further studies were required to show that fasting reduced the false positive rate.

Significant differences in the radiation dose to the patient, usually expressed as the effective dose equivalent (EDE) in mSv can be demonstrated for the various techniques. A 111-Indium labelled white cell scan involving 20MBq of activity gives an EDE of 12 mSv, a uterine absorbed dose of 2mGy and a splenic absorbed dose of 56mGy. A 99m-Tc HMPAO white cell scan involving the administration of 200MBq activity gives an EDE of 3.4 mSv, a uterine-absorbed dose of 0.8 mGy and a splenic absorbed dose of 31.3mGy. A standard barium enema by comparison results in an EDE of 7.69 mSv (range 2.92-33.64mSv), and a uterine/ovarian dose of 16 mGy.[29] This clearly demonstrates the lower radiation dose to the patient associated with 99m-Tc HMPAO white cell scintigraphy in comparison to 111-Indium white cell scintigraphy and conventional barium radiology studies.

TECHNICAL ASPECTS

The search for a 99m-Tc labelled radiopharmaceutical to radiolabel blood cells began in the mid-1970s when McAfee and Thakur tested a number of soluble and colloidal compounds.[1,30] Despite this early work, until the late 1980s, none of the agents tested had given results as good as those obtained with the 111-Indium complexes, 111-Indium oxine and 111-Indium tropolonate. This changed as a consequence of the ground-breaking work of Danpure, Osman and Carroll[31] using the radiopharmaceutical 99m-Tc HMPAO developed by Amersham International principally to allow the study of cerebral blood flow.[18,19] The lipophilic properties of this compound led to interest in it as a potential label for white blood cells. Danpure's group developed the labelling protocol as an *in vitro* technique[32] and then refined it for *in vivo* use, and for the first time produced results comparable to those of 111-Indium labelled compounds.

The technique developed by Danpure *et al.*[31] has been refined over the years, but in most Nuclear Medicine departments which currently perform white cell scanning, the cell labelling technique used is very similar to that first described in 1988. The labelling protocol used in our own department is appended (see Figure 1). The process takes approximately 60 minutes, although samples from up to four different patients can be labelled simultaneously. The technique requires some expertise to avoid white cell activation during the labelling process and to ensure good

1. To a tissue culture flask add 4 ml starch solution, 4 ml saline and 2 ml heparin (2,000 i.u.). Mix gently and coat the internal surface of the flask with the mixture.
2. Mark the 50ml level on the flask with a black line.
3. Use a 19G butterfly needle to remove the blood sample from the patient. With the open end of the butterfly tube in the flask, let the blood drip into the mixture until the 50 ml line is reached. A tourniquet is kept on the patient's arm during this venesection to speed up the blood flow. The blood and anticoagulant starch solution are gently mixed throughout.
4. Place the flask in a laminar flow cabinet in the dispensary and allow the cells to settle for 30 minutes at room temperature.
5. Remove the leucocyte-rich plasma with a sterile pipette and place it in sterile containers for centrifuging. Take off as much as possible without disturbing the red cells.
6. Centrifuge the leucocyte-rich plasma at room temperature for ten minutes at 1,067 rpm.
7. Prepare the 99m-Tc HMPAO while the leucocyte rich plasma is centrifuging. Draw up 800 MBq 1 ml and inject this into the vial of lyophilised HMPAO. Mix gently and allow to stand until leucocyte-rich plasma has been centrifuged. The reconstituted 99m-Tc HMPAO must be used within 30 minutes of preparation.
8. Decant the supernatant platelet-rich plasma into a sterile universal container and keep for preparation of cell-free plasma.
9. Resuspend the white cell pellet in 0.5 ml of sterile saline by gentle mixing.
10. Draw up the 99m-Tc HMPAO and add it to the white cell suspension. Mix gently and incubate at room temperature for ten minutes. Place the container within a lead shield in the laminar flow cabinet.
11. During this ten-minute incubation period, centrifuge the platelet-rich plasma for five minutes at 3,000 rpm. The resultant supernatant is cell-free plasma.
12. After the ten-minute incubation period is complete, add 3 ml of cell-free plasma, mix gently and centrifuge at room temperature for ten minutes at 693 rpm.
13. Pour off supernatant into another bottle. Keep this to measure its activity.
14. Resuspend white cell pellet in a further 3 ml of cell-free plasma as before. Repeat steps 12 and 13 as above.
15. Repeat steps 12 and 13 one further time.
16. Measure the activity of 99m-Tc in the supernatant and the white cell suspension
17. Calculate the % labelling efficiency i.e.

$$\frac{\text{White cell activity} \times 100}{\text{white cell activ.} + \text{supernatant activ.}}$$

18. Draw up the labelled white cell suspension into a 5 ml syringe when the patient is ready. N.B.: The ARSAC limit for this study is 200 MBq of injected activity.
19. The labelled white cells are injected intravenously through a large-bore needle or a butterfly, having ensured that the patient details on the labelled white cell sample correspond to the details on the patient's identification band.
20. Images are acquired at one hour and 2.5 hours post re-injection.

FIGURE 1
Labelling procedure for 99m -Tc HMPAO.

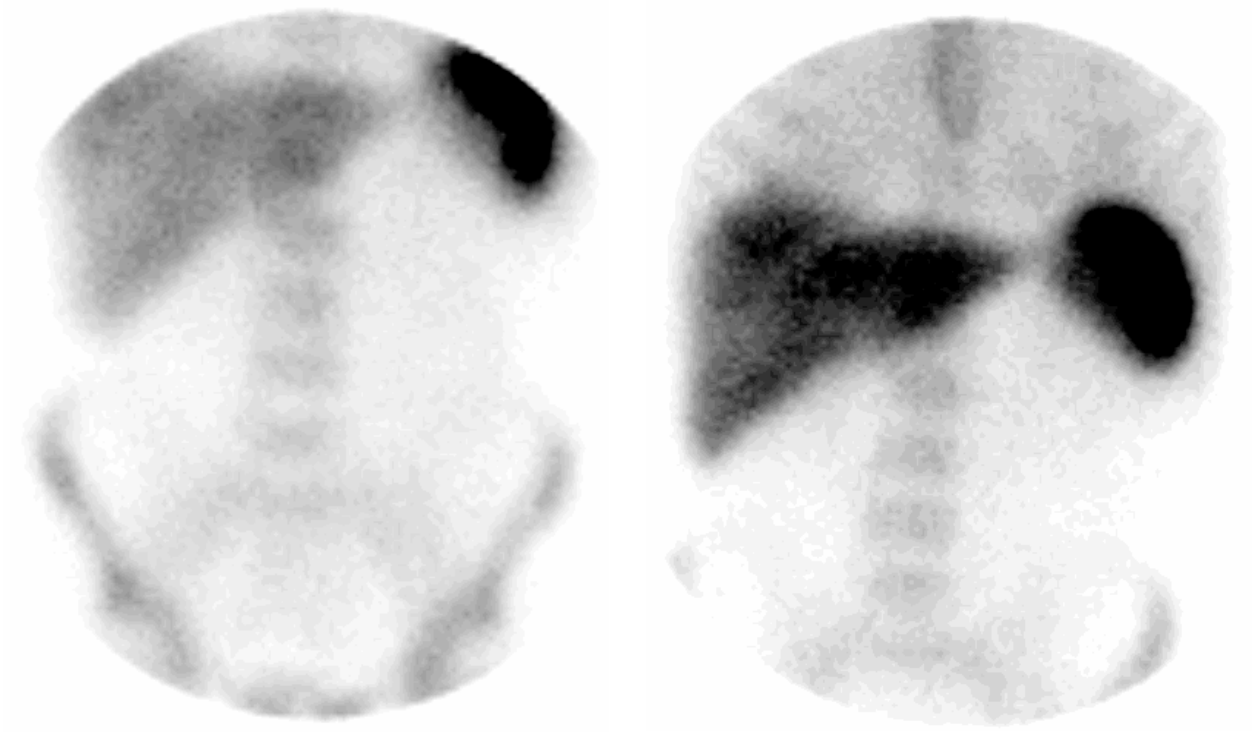

FIGURE 2
A 99m-Tc HMPAO white cell scan.

labelling efficiency. Good aseptic technique is essential, as is very careful labelling of blood samples and patients to avoid errors when the labelled white cells are re-injected. The labelling process occupies one member of technical staff for a full morning, and is usually performed on two or three days per week.

There are no contra-indications to white cell scintigraphy. As with all nuclear medicine investigations, the test should not be performed if at all possible in pregnant women, but this aside, the procedure can be safely and easily applied to all groups of patients. There has been some debate in the nuclear medicine community over the effect of steroids and other immunosuppressive therapy on the results of white cell scintigraphy. In theory, these agents can affect white cell activation and migration as part of their pharmacological action, potentially leading to 'down-grading' of abnormalities on the scans, or even false negative scans. Whilst this has never been studied systematically, there is no evidence to suggest that this is a significant problem in clinical practice.

The average cost of white cell scintigraphy in Glasgow Royal Infirmary is approximately £225 per patient. By comparison, colonoscopy costs about £350 per patient whilst barium studies of the small and large bowel cost £150 and £170 per study respectively. Clearly, not all hospitals contain a nuclear medicine department which is able to offer white cell scintigraphy as a clinical service, and its availability is therefore limited.

IMAGE ACQUISITION

The image acquisition protocol in white cell scintigraphy is dependent to a large extent on the clinical question being posed by the referring clinician. In all cases, two sets of images are acquired. In patients with suspected or proven IBD, the two sets of images are obtained at one hour, and then 2.5-3 hours after re-injection of the labelled white cells. A normal 99m-Tc HMPAO white cell scan is shown in Figure 2. In patients with a suspected intra-abdominal abscess, the images are acquired at one hour and four hours post-reinjection. In this latter group, the delay in the acquisition of the second set of images is to allow greater time for white cell accumulation to occur in the infected/inflammatory focus. In theory this increases the sensitivity of the technique in this situation, but almost certainly does so at the cost of reduced specificity due to increased physiological accumulation of the tracer in the bowel. 111-Indium labelled white cell scintigraphy is superior to 99m-Tc HMPAO scintigraphy for the localisation of intra-abdominal sepsis.[26] This in part reflects the problems with physiological bowel uptake of 99m-Tc HMPAO, and also the advantage which 111-Indium holds over 99m-Tc in relation to its half-life. The significantly longer half-life of 111-In (67 hours), compared to that of 99m-Tc (6 hours), allows delayed imaging at 24 hours or even longer post-re-injection to be performed, enhancing the sensitivity without significantly compromising the specificity. A further disadvantage of 99m-Tc is that by 24 hours, it has decayed to a level which gives unacceptably low target-to-background ratios.

A major drawback of 99m-Tc HMPAO white cell scintigraphy is the number of studies affected by physiological excretion of 99m-Tc HMPAO with increasing time post-re-injection of the labelled cells. Lantto *et al.*[40] studied this effect in a systematic manner and found that at two hours, 7% of studies exhibited non-specific bowel accumulation of the tracer, and that this figure increased to

28% at four hours post-re-injection.

The mechanism of this bowel uptake of the tracer is the subject of some debate. It is extremely unlikely to be delivered by intact leucocytes as the presence of polymorphonuclear leucocytes, the main sub-population labelled, in the intestinal mucosa and lumen are features characteristic of inflammation. To what degree polymorphonuclear leucocytes migrate via the mucosa to the intestinal lumen in normal bowel is unknown but, in order to generate such intensity of 99m-Tc accumulation as is seen in the false positive images, large numbers would be required. Free, unbound pertechnetate is unlikely to be responsible as this is taken up by stomach and thyroid tissue, a finding rarely reported. Nevertheless, the generation of soluble technetium species, as evidenced by urinary excretion, is well recognised after re-injection of 99m-Tc HMPAO labelled white cells. Several groups have noted the relatively frequent finding of gall bladder visualisation during 99m-Tc HMPAO white cell scintigraphy, the incidence of which was increased by fasting in the study of Neilly and Gray. This strongly implicates biliary excretion of the tracer as one of the major sources of physiological accumulation of tracer in the bowel.

In an attempt to reduce the number of false positive or non-diagnostic studies due to physiological accumulation of tracer in the bowel, many Nuclear Medicine departments have adopted a different imaging protocol for white cell scintigraphy performed on subjects with known or suspected IBD. In such patients, images are acquired at one hour and 2.5-3 hours post-re-injection of the labelled white cells. This earlier imaging time for the second set of pictures is an attempt to retain high sensitivity without a reduction in specificity. Despite being widely employed, this protocol has never been prospectively tested in a clinical setting, and to some extent represents a compromise based on the work of Lantto *et al.* outlined above.

Our own department has recently sought to further improve the specificity of white cell scintigraphy by injecting pethidine at the same time as re-injection of the labelled white cells. Pethidine causes spasm of the sphincter of Oddi, and therefore should reduce physiological excretion of the tracer into the gut via the biliary system for the duration of the study. This should reduce the number of 'false positive' studies, with a corresponding increase in the specificity of the technique with no reduction in the sensitivity. Our preliminary results confirm that in those patients to whom pethidine is administered, there is a significant reduction in tracer uptake in the bowel on the delayed images in comparison to a control group who did not receive pethidine. The low dose of pethidine administered (0.3 mg/kg), in conjunction with its short half-life, means that this protocol can be applied to both in-patients and out-patients alike.

CLINICAL ASPECTS

As abdominal pain and alteration of bowel habit can account for a large proportion of consultations in a gastroenterology out-patient clinic, a simple test with a high predictive value for intestinal inflammation would be of enormous benefit. Assessment of the activity and extent of IBD (IBD) can often prove to be problematic. Patients who already have a diagnosis of IBD can present with symptoms suggestive of an exacerbation of their disease, but some of these may have 'post-inflammatory' irritability or dysmotility. Prompt and accurate assessment will permit rapid appropriate treatment of active inflammation with increased doses of aminosalicylic acid drugs or steroids, or prevent such potentially toxic drugs being given to patients with functional bowel disorders.

LIMITATIONS OF OTHER CLINICAL TOOLS

The tools available to the clinician to assess the activity of IBD are relatively limited. Laboratory indicators of the acute phase response, commonly C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR) are often measured. These appear to correlate well with colonic disease, but are less accurate in the assessment of small bowel disease. Overall CRP and ESR measurements respectively have sensitivities of 61% and 60%, and specificities of 83% and 84%. Other acute phase proteins such as orosomucoid, haptoglobin or a-1-antitrypsin have greater predictive values but are not measured routinely.[33-35]

Colonoscopy with biopsy remains the 'gold standard' in the assessment of the activity, and extent of colonic and terminal ileal IBD. However this procedure is not without its limitations. Published rates of reaching the caecum by the colonoscope range between 71% and 96%, however it might be assumed that there is a publication bias in favour of higher rates.[36,37] Factors reported to affect the success of colonoscopy include the experience of the colonoscopist, the age and sex of the patient, and the length of colonoscope used. Terminal ileoscopy is desirable but again not always successful. Complications such as bowel perforation are fortunately rare. Even without these technical limitations, many Gastroenterology Units are facing increasing demands for colonoscopy, and arranging prompt but non-urgent examinations of this variety for the assessment of out-patients is increasingly difficult. In addition, out-patient tolerance of the bowel preparation in the out-patients' clinic can be poor and the examination is often perceived as being uncomfortable. Examination of in-patients may also be limited by the wariness of clinicians of using colonoscopy in suspected acute severe colitis.

The assessment of the extent of bowel involvement is more straightforward in UC in view of the confluent nature of this disease and its localisation to the large bowel. Assessment of small bowel Crohn's disease is however more problematic not only because of its patchy nature, but also because stricturing may limit endoscopic access. An example of a 99m-Tc HMPAO white cell scan demonstrating active Crohn's disease of the terminal ileum and colon is shown in Figure 3. Endoscopic visualisation of the small bowel is possible, however out-patient enteroscopy is limited to an examination of the jejunum and proximal ileum only. Although the full extent of the small bowel can be visualised using the enteroscope with the help of laparoscopy, this is obviously an impractical procedure for routine assessment of IBD. Small bowel radiology remains the most common means of investigating small bowel affected by Crohn's disease. The relative merits of small bowel enteroclysis and follow-through studies are hotly debated.[38] Some suggest that enteroclysis gives greatest mucosal detail, however many patients find the required passage of the enema tube unacceptable, and the simpler but possibly less discriminative barium follow-through examination is often performed as an alternative.

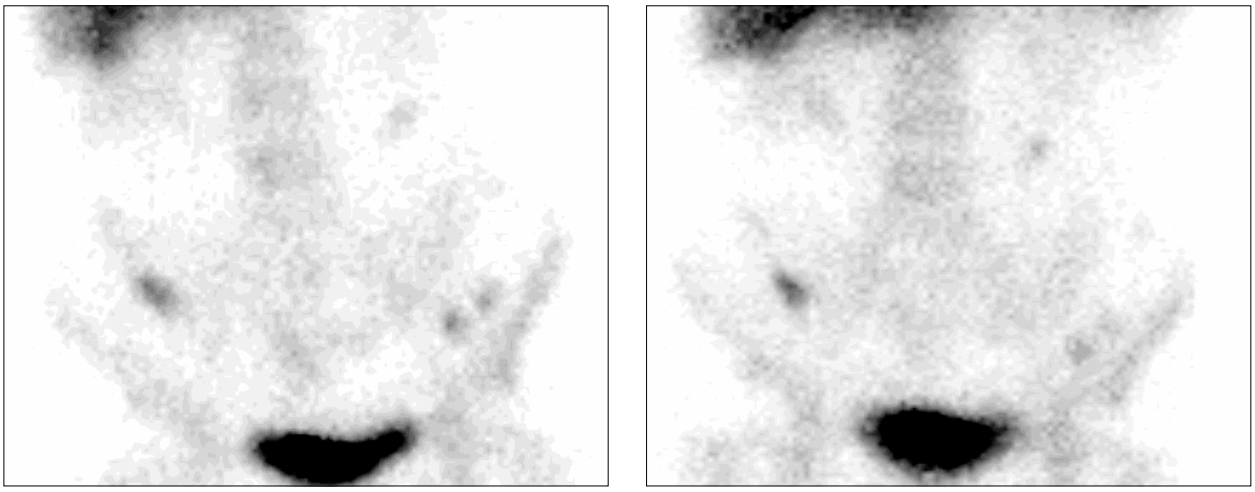

FIGURE 3
A 99m-Tc HMPAO white cell scan demonstrating active Crohn's disease of the terminal ileum and colon.

SCREENING FOR IBD

The diagnosis of IBD should ideally be on the basis of histology from biopsies taken at the time of endoscopic examination. Ulcerative colitis (UC) is usually readily diagnosed at the time of rigid sigmoidoscopy and rectal biopsy. Crohn's Disease however can pose a more difficult problem. Crohn's colitis is patchy perhaps necessitating complete colonoscopy to obtain suitable biopsies. Intubation of the terminal ileum at the time of colonoscopy is achieved less often than reaching the caecum. Isolated small bowel Crohn's disease might be beyond the reach of out-patient enteroscopy and colonoscopy.

White cell scanning has been used to screen patients for IBD. 111-In scanning has been shown to be superior to rectal histology and bowel radiology for this purpose.[6] However 111-In is associated with a significant false negative rate. 99mTc HMPAO scanning seems to be sensitive as 111-In scanning, but has a higher false positive rate. As a screening test 99mTc HMPAO would appear to be more suitable,[25] particularly if the false positive rate can be reduced with the use of pethidine as described above. Therefore white cell scanning has a potential role in the screening of patients and identifying those who might require further investigation. Most clinicians will prefer to obtain histological evidence of IBD if at all possible before labelling a patient with such a diagnosis.

The main role for white cell scanning as a screening tool is probably among children. Ideally paediatric investigation should combine the minimum of discomfort and radiation exposure with a high yield. Relative to endoscopy and barium studies, white cell scanning fulfils these criteria in this age group.[39]

ASSESSMENT OF THE EXTENT OF IBD

If ulcerative colitis is diagnosed and the proximal limit of inflammation can be delineated endoscopically, then further investigation of the extent of disease is not required. However, if initial histology is not able to distinguish ulcerative colitis from Crohn's disease and if there is clinical suspicion of more proximal disease, or if Crohn's disease is demonstrated, then further assessment is indicated. An example of a 99m-Tc HMPAO white cell scan demonstrating ulcerative colitis extending from the rectum to the hepatic flexure is shown in Figure 4.

Leucocyte labelling seems to have a variable specificity and sensitivity for colonic disease (65–100% and 50–100% respectively)[24,40-44] when compared to endoscopic or radiological assessment. Small bowel disease is readily identified on white cell scanning, but difficulty is encountered in differentiating activity in adjacent loops of bowel. In this respect 99mTc HMPAO seems to allow clearer imaging than 111-In scanning. However white cell scanning can only assess the extent of active disease. Chronic quiescent disease could be missed, which if detected might influence the choice of therapy.

ASSESSMENT OF IBD ACTIVITY

Bowel uptake of labelled white cells correlates well with clinical and histological measurements of IBD activity. Measurement of the faecal excretion of 111-In labelled leucocytes over four days has a greater sensitivity than 111-In imaging. Faecal collection however delays assessment and requires the patient to co-operate; the absence of significant constipation is also important. 99mTc HMPAO imaging correlates well with faecal excretion of 111-In labelled leucocytes.[43] Such imaging is effective in separating active from inactive IBD with a sensitivity of 96% and a specificity of 98%.[45] In addition, the response to treatment can also be monitored, as a majority of patients will demonstrate a reduction in the intensity of bowel uptake with clinical remission.

It is a procedure which can be performed safely even with patients who have acute severe colitis and in this respect it potentially has a great advantage over barium contrast radiology or colonoscopy. However in this situation the clinical signs are usually clear, and it is often on the basis of clinical signs or plain abdominal radiology that management decisions are made.

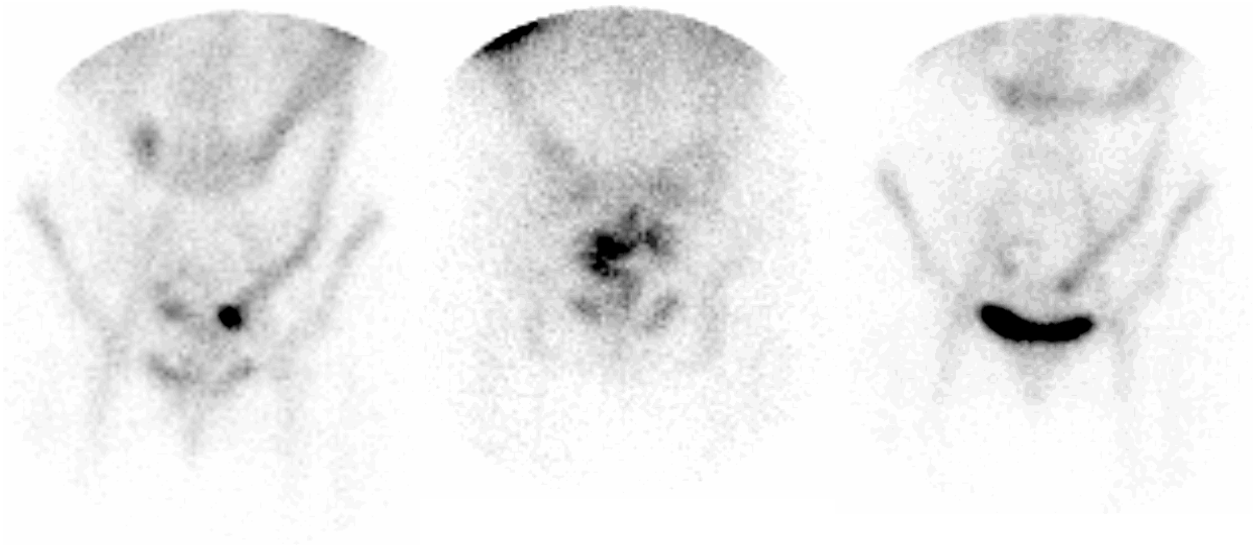

FIGURE 4
A 99m-Tc HMPAO white cell scan demonstrating ulcerative colitis extending from the rectum to the hepatic flexure.

ASSESSMENT OF IBD COMPLICATIONS

Some of the complications of Crohn's disease might be identified by white cell scanning. Intra-abdominal abscesses are identified by persistent uptake seen on delayed images. In one study 111-In scanning was more sensitive in this respect than ultrasound examination; 99mTc HMPAO has similarly been shown to be effective in detecting abscesses and can differentiate them from diverticulitis.[46] White cell scanning probably has little to add to radiology, endoscopy or clinical examination in the other complications of Crohn's disease such as fistulae, strictures and perianal disease. However the presence of a fibrous stricture requiring surgery might be hinted at by symptoms of intermittent bowel obstruction without any evidence of active inflammation on white cell scanning. Alternatively a stricture with intense uptake on white cell scanning might imply a predominantly inflammatory lesion prompting intensive medical therapy before resorting to surgery.

CONCLUSIONS

In practice the assessment of IBD is usually through a combination of clinical, radiological, endoscopic, and histological evidence. White cell scanning gives the clinician another string to their bow. It has obvious limitations in diagnosis, but it probably has a role as a screening procedure for the large number of first-time patients attending with altered bowel habit and abdominal pain. It permits assessment of areas of the bowel inaccessible to routine endoscopy and so may aid to determine the extent of disease. However its main use is probably in the assessment of patients with established IBD who present with a symptomatic apparent exacerbation of their disease. White cell scanning can allow rapid accurate assessment of disease activity and direct therapeutic intervention, and possibly allow response to be monitored.

REFERENCES

1 McAfee JG, Thakur ML. Survey of radioactive agents for *in vitro* labelling of phagocytic leucocytes. I Soluble agents *J Nuc Med* 1976; **17:**480-7.

2 Saverymuttu SH, Peters AM, Hodgson HJ *et al*. Indium 111 autologous leucocyte scanning: comparison with radiology for imaging the colon in IBD. *BMJ* 1982; **285:**255-7.

3 Saverymuttu SH, Camilleri M, Rees H *et al*. Indium 111 Granulocyte scanning in the assessment of disease extent and activity in IBD. *Gastroenterology* 1985; **90:**1121-8.

4 Coleman RE, Black RE, Welch DM, Maxwell JG. Indium 111 labelled leucocytes in the evaluation of suspected abdominal abscesses. *Am J Surg* 1980; **139:**99-104.

5 Saverymuttu SH, Keshavarzian A, Gibson R *et al*. Hepatic abscesses associated with Crohn's disease detected by Indium 111 leucocyte scanning. *J Clin Gastroenterol* 1985; **7:**273-6.

6 Saverymuttu SH, Peters AM, Crofton ME *et al*. 111-Indium granulocytes in the detection of IBD. *Gut* 1985; **26:**955-60.

7 Danpure HJ, Osman S, Brady F. The labelling of blood cells in plasma with Indium-111 tropolonate. *Br J Radiol* 1982; **55:**247-9.

8 Segal AW, Ensell J, Munro JD, Sarner M. Indium-111 tagged leucocytes in the diagnosis of IBD. *Lancet* 1981; **2:**230-2.

9 Saverymuttu SH, Peters AM, Lavender JP *et al*. Indium 111 autologous leucocytes in IBD. *Gut* 1983; **24:**293-9.

10 Carpani de Kaski M, Peters AM, Knight D *et al*. Indium 111 Whole body retention: a new method for quantification of disease activity in IBD. *J Nuc Med* 1992; **33:**756-62.

11 Wistow BW, Grossman ZD, McAfee JG *et al*. Labelling of platelets with oxine complexes of Tc-99m and Indium-111. Part 1-*In vitro* studies and survival in the rabbit. *J Nuc Med* 1978; **19:**483-7.

12 Linhart N, Bok B, Gougerot M *et al*. Tc-99m labelled human leucocytes. an *in vitro* functional study. *Acta Haematol* 1980; **63:**71-80.

13 Kelback H, Fogh J. Technetium 99m labelling of polymorphonuclear leucocytes: preparation with two different stannous agents. *J Nuc Med* 1985; **26:**68-71.

14 Scroth HJ, Oberhausen E, Berberich R. Cell labelling with colloidal substances in whole blood. *Eur J Nuc Med* 1981; **6:**469-72.

15 Danpure HJ, Osman S, Saverymuttu SH *et al*. Evaluation of Tc-99m tin colloid as a selective in vitro labelling agent for

phagocytic cells in whole blood. *Br J Radiol* 1985; **58:**922.
16 McAfee JG, Subramanian G, Gagne G. Technique of leucocyte harvesting and labelling: Problems and perspectives. *Sem Nuc Med* 1984; 14:83-106.
17 Peters AM, Osman S, Henderson BL *et al*. Clinical experience with 99m-Tc Hexamethylpropyleneamineoxime for labelling leucocytes and imaging inflammation. *Lancet* 1986; ii 946-9.
18 Nowotnik DP, Canning LR, Cumming SA *et al*. Development of a Tc-99m labelled radiopharmaceutical for cerebral blood flow imaging. *Nuc Med Commun* 1985; **6:**437-41.
19 Ell PJ, Hocknell JML, Jarritt PH *et al*. A Tc-99m labelled radiotracer for the investigation of cerebral vascular disease. *Nuc Med Commun* 1985; **6:**437-41.
20 Peters AM, Roddie ME, Danpure HJ *et al*. Tc-99m HMPAO labelled leucocytes: comparison with Indium-111 tropolonate labelled granulocytes. *Nuc Med Commun* 1988; **9:**449-65.
21 Li DJ, Middleton SJ, Wraight EP. Tc-99m and Indium-111 leucocyte scintigraphy in IBD. *Nuc Med Commun* 1992; **13:**867-70.
22 Allan RA, Sladen GE, Bassingham S *et al*. Comparison of similtaneous Tc-99m HMPAO and Indium-111 oxine labelled white cell scans in the assessment of IBD. *Eur J Nuc Med* 1993; **20**(3):195-200.
23 Kennan N, Hayward M. Tc-99m HMPAO labelled white cell scintigraphy in Crohn's disease of the small bowel. *Clin Radiol* 1992; **45:**331-4.
24 Scholmerich J, Schmidt E, Schumichen C *et al*. Scintigraphic assessment of bowel involvement and disease activity in Crohn's disease using Tc-99m HMPAO as a leucocyte label. *Gastroenterology* 1988; **95:**1287-93.
25 Gibson P, Lichtenstein M, Salehi N *et al*. Value of positive technetium 99m leucocyte scans in predicting inflammation. *Gut* 1991; **32:**1502-7.
26 Mountford PJ, Kettle AG, O'Doherty MJ, Coakley AJ. Comparison of technetium 99m-HMPAO leukocytes with indium 111 oxine leukocytes for localising intra-abdominal sepsis. *J Nuc Med* 1990; **31:**311-5.
27 Costa DC, Lui D, Ell PJ. White cells labelled with 111-In and 99m-Tc - a study of relative sensitivity and in vivo viability. *Nuc Med Commun* 1988; **9:**725-31.
28 Neilly JB, Gray HW. Gall bladder visualisation in 99m-Tc HMPAO autologous white cell scintigraphy. *Nuc Med Commun* 1996; **17:**243-5.
29 Shrimpton PC, Wall BF, Jones DG *et al*. National survey of doses to patients undergoing a selection of routine X-ray examinations in English hospitals. NRPB R200.
30 McAfee JG, Thakur ML. Survey of radioactive agents for in vitro labelling of phagocytic leucocytes. II Particles. *J Nuc Med* 1976; **17:**488-92.
31 Danpure HJ, Osman S, Carroll MJ. The development of a clinical protocol for the radiolabelling of mixed leucocytes with 99m-Tc hexamethylpropyleneamine oxime. *Nuc Med Commun* 1988; **9:**465-75.
32 Danpure HJ, Osman S, Carroll MJ. *In vitro* studies to develop a clinical protocol for radiolabelling mixed leucocytes with 99m Tc HMPAO. *Nuc Med Commun* 1987; **8:**280.
33 Calvin J, Neale G, Fotherby K *et al*. The relative merits of acute phase proteins in the recognition of inflammatory conditions. *Ann Clin Biochem* 1988; **25:**60-6.
34 Fagan E, Dyck R, Maton P *et al*. Serum levels of C-reactive protein in Crohn's disease and ulcerative colitis. *Eur J Clin Invest* 1982; **12:**351-9.
35 Weldon MJ, Lowe C, Joseph AEA, Maxwell JD. Review article: quantitative leucocyte scanning in the assessment of IBD activity and its response to therapy. *Ali Pharm Ther* 1996; **10:**123-32.
36 Dijkstra J, Reeders JW, Tytgat GN. Idiopathic IBD: endoscopic and radiologic correlation. *Radiology* 1995; **43:**124-6.
37 Saunders BP. Viewing the colon: imaging and colonoscopy. *Medicine* (Gastroenterology) 1998; **26:**53-6.
38 Maglinte DDT, Kelvin FM, Lappas JC *et al*. Current state of small bowel radiography. *Abdom Imaging* 1996; **21:**247-57.
39 Shah DB, Cosgrove M, Rees JL, Jenkins HR. The technetium white cell scan as an initial imaging investigation for evaluating suspected childhood IBD. *J Ped Gastro Nut* 1997; **25:**524-8.
40 Lantto E, Jarvi K, Lantto T, Taavitsainen *et al*. Technetium-99m hexamethyl propylene amine oxime leucocytes in the assessment of disease activity in IBD. *Eur J Nuc Med* 1993; **20:**227-36.
41 Papos M, Nagy F, Lang J, Csernay L. Technetium-99m hexamethyl propylene amine oxime labelled leucocyte scintigraphy in ulcerative colitis and Crohn's disease. *Eur J Nuc Med* 1993; **20:**766-9.
42 Spinelli F, Milella M, Sara R, Banfi F *et al*. The 99mTc HMPAO leucocyte scan: an alternative to radiology and endoscopy in evaluating the extent and activity of IBD. *J Nuc Biol Med* 1991; **35:**82-7.
43 Mansfield JC, Gaiffer MH, Tindale WB, Holdsworth CD. Quantitative assessment of overall IBD activity using labelled leucoytes: a direct comparison between indium-111 and technetium-99m HMPAO methods. *Gut* 1995; **37:**679-83.
44 Gaiffer MH. Labelled leucocyte scintigraphy in IBD: clinical applications. *Gut* 1996; **38:**1-5.
45 Becker W, Fischbacher W, Weppler M *et al*. Radiolabelled granulocytes in IBD: diagnostic possibilities and clinical indications. *Nuc Med Commun* 1988; **9:**693-701.
46 Weldon MJ, Joseph AE, French A *et al*. Comparison of 99m technetium hexamethylpropylene-amine oxime labelled leucocyte scanning and ultrasound in the diagnosis of intra abdominal abscess. *Gut* 1993; **37:**557-64.