

Are Leukocytes Labeled with Stabilized ^{99m}Tc -HMPAO Becoming Activated During Labeling?

TO THE EDITOR: We read with interest the recent article by Robins et al. (1) in which they reported biokinetic data of leukocytes labeled with stabilized ^{99m}Tc -HMPAO (exametazime) by a method that had earlier been described by their group in this journal (2) and elsewhere (3), with minor differences in methodology and results.

Labeling white cells with technetium rather than indium has obvious advantages in terms of availability, image quality, and radiation absorbed dose. It became a clinical reality with the introduction of HMPAO, first described for white cell labeling by Peters et al. (4) in 1986, and has since proved useful in imaging acute inflammation and inflammatory bowel disease. Hung et al. (2) have aimed to take this further by using HMPAO stabilized with methylene blue. This, however, requires a change in cell isolation procedure, because the dark blue color makes it impossible to distinguish the leukocyte sediment from the supernatant. Hung et al. have suggested using a double-dilution technique. Their protocol requires the addition of a total of 17 mL of a 12.6% acid citrate dextrose (ACD) solution in normal saline.

The biokinetics described by Robins et al. (1) can be summarized as follows. Recovery at 30 min and 1 h was 18% and 16%, respectively. The ratio of activity in liver to that in spleen changed from 1.3 at 1 h to 1.6 at 24 h after injection. Residence time in the lung was 0.55 h, and the mean half-clearance time was 16 min.

We are concerned that these biokinetics may represent a mild-to-moderate form of white cell activation during labeling. Robins et al. (1) already pointed out that the half-clearance time in the lung in their study was longer than the previously reported times of 7.7 min for Becker et al. (5) and 9.8 min for Brown et al. (6). We do not agree with the observation of Robins et al. that there was no visual evidence of prolonged lung retention. In Figure 2 in their paper, considerable lung uptake can still be seen 8 h after injection, which one would expect to observe only in a 1-h image. A slight progressive sequestration in the liver (rather than physiologic pooling in the spleen) and the surprisingly low recovery (which is normally in the 30%–40% range, again with no differences between technetium- and indium-labeled leukocytes), are also indicative of cell activation.

Indeed, higher than normal lung uptake with a prolonged intrapulmonary transit time, followed by a progressive accumulation in the liver rather than the spleen, are clear signs of white cell activation during labeling (5). There is still no better method for evaluating the viability of radiolabeled white cells than studying their kinetics in vivo.

Apart from general manipulation during the cell isolation and labeling process, there are two possible causes for leukocyte activation

in this study: the labeling in saline rather than plasma and the methylene blue buffer solution.

The former has been extensively investigated in connection with the question of whether white cell labeling with indium should be performed with oxine (in saline) or tropolone (in plasma). There is a general consensus that labeling leukocytes in plasma rather than saline is advantageous because the cells are maintained in their normal physiologic environment. Labeling white cells in saline has long been recognized as a possible cause of cell activation, although clinical studies on the preference of oxine or tropolone gave conflicting results.

The influence of the latter factor, methylene blue, on leukocyte activation is not known. It can, however, indirectly be determined by changing the cell isolation protocol such that the ACD–saline solution is replaced with plasma, which can easily be achieved by moderately increasing the amount of blood drawn. However, labeling in plasma is known to lead to a decrease in labeling efficiency (7). Because stabilized HMPAO provides a remarkably high labeling efficiency in saline (1), it might be expected to give an acceptably high labeling efficiency in plasma as well. If the biokinetics of the white cells labeled with stabilized HMPAO in plasma do not correspond to what has previously been described, the methylene blue buffer solution is likely to have a detrimental effect. If it does, the use of stabilized HMPAO will be a welcome contribution to the daily work of any nuclear medicine department performing white cell labeling in vitro.

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