Quality Control of Radiolabeled Leukocytes with Monoclonal Antibodies

TO THE EDITOR: A recent letter by Deborah A. Kaminsky in the May 1993 issue of the *Journal* highlights the importance of leukocyte (WBC) purification prior to labeling for autologous leukocyte imaging of infection (*1*). Traditional methods for WBC purification fail to adequately reduce the number of contaminating red blood cells (RBC) and platelets which may lead to false-positive studies or high background levels (*2*).

One method of removing excessive RBC is by hypotonic lysis. This method has been used and results indicate that viable and functional WBC populations are capable of producing reliable images of infected foci when labeled with ^{99m}Tc-HMPAO (*3*).

We have been studying the kinetics of WBC during separation and labeling with ^{99m}Tc-HMPAO by monitoring the expression of activation antigens CD11b and CD18 (*4*) during various stages of purification and labeling.

Results indicate that hypotonic lysis leads to a high degree of neutrophil activation and alters the relative proportions of the leucocytes by eliminating a significant percentage of the lymphocytes (Tables 1 and 2).

TABLE 1
Differential WCC During Separation and Labeling with ^{99m}Tc-HMPAO

Step no.	Differential WCC		
	%Neutrophil	%Lymphocyte	%Monocyte
1 Whole blood	65.9 ± 11.2	22.5 ± 9.4	7.4 ± 3.3
2 After sedimentation	64.9 ± 9.5	22.8 ± 9.8	7.4 ± 3.4
3 After RBC lysis	75.3 ± 1.5	11.0 ± 2.0	8.7 ± 3.2
4 After ^{99m}Tc-HMPAO	74.8 ± 7.2	12.4 ± 5.9	8.5 ± 3.2

Mean and standard deviation of eight patients.
Separation and labeling kinetics of WBCs with ^{99m}Tc-HMPAO.
RBC lysis is achieved by a 20-sec hypotonic lysis followed by re-buffering and washing.

TABLE 2
Neutrophil Activation During Separation and Labeling with ^{99m}Tc-HMPAO

Step no.	Activation index
1 Whole blood	1.00 ± 0.00
2 After sedimentation	1.09 ± 0.15
3 After RBC lysis	2.25 ± 0.94
4 After ^{99m}Tc HMPAO labeling	2.34 ± 1.17

Mean and standard deviation of eight patients.
Activation index = 0.5 (CD11b + CD18) − CD13.
Mean channel fluorescence normalized to whole blood value.
CD13 = Control anti-granulocyte monoclonal antibody.

Although we did not observe any loss in viability with trypan blue, the degree of activation caused by lysis may alter the functional capabilities of WBC, which could lead to variable results when interpreting the nuclear scans (*5*).

Monoclonal antibody activation studies appear to be useful in the quality control of leukocyte separation and labeling procedures.

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Effects of Erythromycin on Alcohol Absorption

TO THE EDITOR: We congratulate Edelbroek et al. (*1*) on a carefully done study relating increased gastric emptying due to erythromycin to an increase in the apparent absorption (systemic exposure) of alcohol, which they attribute in part to reduced metabolism of alcohol by the gastric mucosa. However, we take issue with this proposed mechanism and offer an alternative interpretation of their data.

Edelbroek et al. (*1*) found an inverse correlation between peak blood alcohol concentration (Cmax) and 50% emptying time of liquid during the control period but not the erythromycin period. Inspection of the data strongly suggests that all of the points from both study periods fall along a single regression line; estimation of the values from the graph indicates that the combined r value may be as great as −0.83, implying that up to 69% of the variability in Cmax can be explained simply by differences in gastric emptying. We have recently obtained similar results in a study of the effects of famotidine on the apparent absorption of alcohol (*2,3*). Changes in Cmax induced by famotidine correlated strongly, $r = -0.62$, $p = 0.001$, with changes in the time at which Cmax was achieved (Tmax), with Tmax being an indirect estimate of the liquid emptying rate. Mean plasma ethanol Cmax was increased 23% relative to control ($p = 0.013$) in 24 male subjects when famotidine (40 mg) was given with a standard breakfast 1 hr prior to a small oral ethanol dose (0.15 g/kg).

Recently, it has been suggested that there is no need to attribute changes in alcohol absorption induced by H_2-receptor antagonists to alterations in the activity of gastric alcohol dehydro-