LETTERS TO THE EDITOR

Detrimental Effect of Indium-111 on Human Lymphocytes?

In a recent issue of the *Journal*, ten Berge et al. concluded that indium-111 "has detrimental effect on human lymphocytes" (*1*). This article must certainly be viewed with apprehension, since several centers are using indium-111 labeling techniques. As we are also involved in the study of indium-111 cytotoxicity, we would like to question ten Berge's data and their conclusions.

According to ten Berge, lymphocyte proliferation was inhibited by doses of indium-111 greater than 2 μCi for 10^7 cells. We have found that even larger doses do not affect lymphocyte phenotypic expression as evaluated by monoclonal antibodies (*2*) and that the cells are capable of recirculating and migrating into peripheral organs (*3*). As far as chromosomal aberrations induced by indium-111 are concerned, the data published by ten Berge included only two subjects. In one of them, the reported cellular spontaneous aberrations (i.e., cells not incubated with indium-111) were 14%, whereas the accepted normal level of spontaneous aberrations is no more than 3%. In our opinion this subject should have not been included. Furthermore, using 3 μCi $\times$ 10^7 cells, ten Berge observed over 50% chromosomal aberrations. As 3 μCi $\times$ 10^7 cells correspond only to 85 rad for 48 hr exposure, or 128 rad for 72 hr exposure—the culture times used by ten Berge—even without allowing for the radioactive decay (*4*), this finding is in contrast with data previously produced by one of the coauthors (*5*), indicating that lymphocyte irradiation with 100 rad induced acentric and dicentric aberrations below 20%.

Last but not least, since it has been shown that "there is not one human radiosensitivity to chromosomal aberrations production but, even amongst normal donors, there exist great differences" (*5*), we doubt whether on the basis of one single subject, ten Berge et al. can claim that "indium-111 has detrimental effect on human lymphocytes."

Further studies on a much larger number of subjects are necessary before drawing any conclusion on indium-111 cytotoxicity.

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Reply

In reply to the letter of Pozzilli et al., I would like to make the following comments regarding the data on chromosome aberrations.

1. Irrespective of the frequencies of spontaneous aberrations, lymphocytes from both the subjects responded with a dose-dependent increase in chromosomal aberrations on incubation with In-111, which indicates that In-111 induces chromosomal aberrations.

2. The comparison of the induced frequencies of aberrations by In-111 and x-rays is not totally valid, as the treatment regimes are different. X-irradiation is done during G_0, a relatively radioresistant cell stage, whereas In-111 was present during all the stages of cell cycle, some of which are extremely radiosensitive. In addition, there are the concepts of RBE and LET in the radiobiological effects of different radiations, and Pozzilli et al. do not seem to be aware of this.

3. The interindividual variability for radiosensitivity is within a factor of two, and there is no really great difference. Irrespective of the inherent sensitivity, there is always a good dose response, which is the case with the two subjects used in our study.

I suggest that the physicians should use the concepts of "benefit-risk assessment" for the patients before using In-111-labeled lymphocytes.

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Re: First-Pass Measurements of Regional Blood Flow with External Detectors

In a recently published article, Drs. Mullani and Gould presented a first-pass model to measure blood flow (*1*). Using peak-counts method, they applied the model to the measurement of myocardial blood flow using rubidium-82 as the tracer. An excellent linear correlation between the blood flow measured by their method and by the labeled-microsphere method was seen, indicating the general validity of the peak-counts method. The slope of the relationship deviated from the ideal slope of unity, however, indicating a systematic error in the method. I would like to suggest a source for this error.

The first-pass model requires that the sampling time be shorter than the shortest transit time of the tracer through the capillary bed in the region of interest. This shortest transit time can be measured by rapid sequential venous sampling after bolus arterial

injection. These data are readily available from studies that evaluate the extraction of tracers by different organs. For coronary circulation, the shortest transit for rubidium, as well as for other incompletely extracted tracers, is less than 8 sec (*2,3*). Rubidium is seen in the earliest venous sample in which the strictly intravascular tracer appears, indicating that part of the rubidium is intravascular during the first pass. Thus, the shortest transit time for rubidium is the same as that of any intravascular tracer.

The first-pass model using the peak-counts method as proposed by the authors requires that the total first-pass input of activity to that region (of interest) be under the view of the detector prior to any washout. This requirement is related to the shortest transit time. In both myocardial and cerebral studies using external detector and different tracers injected as extremely short arterial boluses, the washout of the tracers from the region of interest is seen at less than 8 sec postinjection, indicating that the input bolus must have a duration of less than 8 sec for the peak-counts method to be valid (*4,5,6*). Yet Mullani and Gould show that the input bolus has a much longer duration (Fig. 2, p 579). Measured from the time when the arterial concentration reached 2000 counts (1/9 of maximum) to when the activity declined to 2000 counts, the bolus duration is 12–13 sec. Therefore, venous outflow must have occurred before cessation of arterial input and their sampling time of t_m. This leads to a systematic underestimation of blood flow by the peak-counts method with corresponding decrease of the slope from the ideal slope of unity.

This requirement for the short duration of input bolus places a limitation on the applicability of the first-pass model. Since in most cases an arterial injection is required to achieve this short bolus, the intravenous applicability of this method is quite limited.

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Reply

The first-pass model for regional blood flow (*1*) is based on the assumption that during the first several seconds of the transit of a bolus of tracer through a region, there exists a minimum transit-time delay, t_d, during which the tracer is entering the region but has not begun to leave it. During that time therefore, the venous egress from the region is zero and can be ignored to yield the following equation for measuring regional blood flow:

$$F = \frac{P(T)}{\int_0^T C_a(t)dt}, \qquad (1)$$

where, P(T) is the detected count rate from the region of interest, $C_a(t)$ is the arterial concentration, F is the flow, and T is less than t_d.

[Note: In the original paper, $\bar{t}$ was used to indicate the minimum transit-time delay (TTD). In this manuscript, $\bar{t}$ is replaced by t_d to remove the possibility of mistaking the minimum transit-time delay for the mean-transit time defined by V/F (*4*).]

As long as the detection time, T, is smaller than the minimum transit-time delay, t_d, regional blood flow can be measured in any region regardless of whether the tracer is extracted or not. In theory, this model is valid except when the transit time becomes extremely small.

The first-pass model of regional blood flow was modified to a peak-counts model by using the properties of the mass balance equation, since at the peak-count time, t_m, the rate of change of the detected count rate is equal to zero. At the peak, therefore, the rate of input of the tracer from the arterial side equals the rate of its exit through the venous side. The peak-counts model has several advantages, as described by Mullani and Gould in their manuscript. However, as Yen (*2*) points out in his letter to the editor, the peak-counts model may be of limited application in those situations in which the venous egress may not be negligible at the time of peak counts.

The errors in blood-flow measurements by the peak-counts model will depend on the bolus size, bolus duration, the minimum transit time of the bolus through the region of interest, and the extraction of the tracer. This subject needs to be studied experimentally in greater detail before exact determination can be made on the limitations of the peak-counts model. However, in the absence of any published data on the regional characteristics of transit times, we wish to present some of our preliminary data (unpublished) in order to suggest that the errors may not be large and to clarify the problem to the readers. These preliminary data will also indicate where future experimental data are needed.

The effect of bolus size on the measurement of extraction fraction and blood flow was evaluated using the first-pass extraction fraction method of Mullani et al. (*3*). Various volumes of rubidium-82 chloride were injected in the femoral vein of a dog and the myocardial count rate was measured with a beta probe over the heart. Bolus injections ranging from 1 to 20 cc were made in less than one second for each injection followed by a 10-cc saline flush. There was very little difference in the measurement of blood

TABLE 1. EFFECT OF BOLUS SIZE ON ESTIMATION OF FIRST-PASS EXTRACTION FRACTION OF RUBIDIUM-82 AND BLOOD FLOW IN MYOCARDIUM

Bolus size (cc)	Extraction fraction	Regional blood flow (ml/min-g)
1	0.32	2.5*
2	0.383	2.08
5	0.363	1.96
10	0.384	1.99
20	0.384	2.01

* The 1 cc bolus injection was statistically poor due to small amount of radiotracer injected.