lowed by incubation with pertechnetate, causes $^{99m}$Tc to bind to the red blood cells even when no residual tin could be found in the red cells. In our own unpublished experiments, the efficiency of $^{99m}$Tc binding to red cells increased with the time of exposure of the red cells to tin or $^{99m}$Tc. We originally favored the hypothesis that Sn(II) entered the cells and awaited the entry of pertechnetate, which it subsequently reduced from its anionic (VII) state to the cationic (IV) state, in which form the technetium was then chelated by intracellular electronegative sites (*3*). However, a recent article in *Science* (*4*) has shown that tin is an unusually potent inducer of heme oxygenase in the kidney. Such induction was rapid and significant, and it occurred at levels of administered Sn(II) within an order of magnitude of the Sn(II) dose found in some radiopharmaceutical preparations and at substantially lower levels than that found in some diets. Thus, it seems reasonable to hypothesize that Sn(II) acts indirectly by altering redox mechanisms in the choroid plexus and red cells, resulting in a selective in situ capacity for reducing Tc(VII) to Tc(IV). Should this prove to be the case, it may be that the rate of Tc(VII) reduction (or reduction of similar metallic anions), as evidenced by regional fixation of selected radionuclides, may prove to be a sensitive measure of certain alterations in in situ redox systems. Practitioners of nuclear medicine should attempt to determine whether such Sn(II)-induced in situ redox changes, measured in vivo with radiopharmaceuticals, can offer new insights into pathophysiologic processes.

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## Preparation of $^{99m}$Tc-Labeled Red Blood Cells

The paper by Smith and Richards (*1*) deserves praise for its thoroughness in determining the conditions for $^{99m}$Tc labeling of red blood cells (RBCs) with stannous citrate. We examined these parameters and found that 3–8 μg of stannous ion per 10 ml of whole blood is effective when stannous glucoheptonate is used to prepare $^{99m}$Tc-labeled RBCs. Based on this observation, we have modified the stannous glucoheptonate method (*2*) to require 7% of the reported amount of stannous glucoheptonate (equivalent to 4 μg of stannous ion and 14 mg of sodium glucoheptonate), and we have eliminated the EDTA and one saline wash. In a further simplification of this method, we substituted a syringe apparatus (*3*) for the Unitag Bag serving as the tagging vessel. Labeling efficiencies in excess of 95% are obtained with canine and human RBCs. The time and number of centrifugations are comparable to the authors' method, and all the necessary components are commercially available in sterile pyrogen-free form.

We emphasize the lack of commercial availability of the authors' lyophilized stannous citrate kit, and the considerable effort, expertise, and equipment required to prepare these kits, compared to the readily available stannous glucoheptonate. These facts make it difficult to agree with the authors' statement that the stannous glucoheptonate method (either as published or as modified) is much more involved than the lyophilized stannous citrate method.

We note the remarkable splenic uptake of heat-damaged (HD) $^{99m}$Tc-labeled RBCs reported in patients (90%). However, the specific method by which this figure was determined is not presented. Smith and Richards hypothesize that high splenic uptake is dependent on the injection of "small" volumes of HD radiolabeled RBCs, but the volumes injected are not reported. Working on the assumption that a 70-kg patient would receive 0.2 ml of HD $^{99m}$Tc-RBCs, we determined the volume of packed RBCs to be 0.0029 ml per kilogram. This calculation was based on the authors' reported specific activity of greater than 15 mCi/ml RBCs (*4*) and assuming an injected dose of 3 mCi of HD $^{99m}$Tc-RBCs. In order to examine their hypothesis, we injected seven rats with homologous HD $^{99m}$Tc-RBCs with the following volumes of packed RBCs: 0.002 ml/kg (two rats); 0.02 ml/kg (two rats); 0.2 ml/kg (two rats); 2.0 ml/kg (one rat). The animals were killed 1 hr after intravenous injection and spleen and carcass radioactivity were measured. No correlation was observed between splenic uptake and volume of packed RBCs. For the seven rats, the splenic uptake was 49.1 ± 4.4% (mean ± s.d.) of the injected dose, which is similar to our earlier published results (*2*).

The authors do not explain the reason for the apparent increase in splenic uptakes from 66% in their previously published report (*4*) to the 90% figure in their recent article (*1*).

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## Reply

As we would with any radiopharmaceutical, we welcomed the improvements by Gutkowski et al. on their stannous glucoheptonate RBC-labeling method. We recognize that, while all the components required for their method are commercially available, the system is not available as a unit kit. We had not expected that individual investigators would