The purpose of this study was to evaluate the role of both protein and radionuclide in the accumulation of \( {^{111}}\text{In-labeled human immunoglobulin G} \) (IgG) in infectious foci. In rats with a calf muscle infection, biodistribution was determined 2, 6, 24, and 48 hr after injection of a radiopharmaceutical. For IgG, human serum albumin (HSA) and human immunoglobulin A (IgA), all labeled with \( {^{111}}\text{In} \), target-to-background (T/B) ratios were similar throughout the study. However, absolute absorbance of \( {^{111}}\text{In-IgA} \) was significantly lower. For IgG labeled with \( {^{111}}\text{In}, {^{123}}\text{I}, \) or \( {^{99m}}\text{Tc} \), similar T/B ratios were found up to 24 hr. After 48 hr, the T/B ratio of \( {^{111}}\text{In-IgG} \) was significantly higher than the T/B ratio of \( {^{123}}\text{I-IgG} \). The absolute absorbance of \( {^{111}}\text{In-IgG} \) was higher than that of \( {^{99m}}\text{Tc-IgG} \) at 24 hr and \( {^{123}}\text{I-IgG} \) at 48 hr. In conclusion, the radio­nuclide appears to be of major importance in the accumulation of radiolabeled proteins in infectious foci. Protein mainly influences blood clearance and distribution in organs. The Fc–γ receptor is not crucial for accumulation in infectious foci.


Several reports suggest the utility of scintigraphy with \( {^{111}}\text{In-labeled nonspecific polyclonal human immunoglobulin G} \) (\( {^{111}}\text{In-IgG} \)) for the detection of various types of focal infection in humans (1–4). However, the mechanism of \( {^{111}}\text{In-IgG} \) accumulation in infectious and noninfectious inflammatory foci is not fully understood. Both specific receptor interaction and nonspecific macromolecular entrapment have been proposed (5–8). To evaluate the role of the type of protein in the mechanism of \( {^{111}}\text{In-IgG} \) accumulation in infectious foci, \( {^{111}}\text{In-IgG} \) was compared to \( {^{111}}\text{In-labeled human serum albumin} \) (HSA), and \( {^{111}}\text{In-labeled human immunoglobulin A} \) (IgA); also \( {^{123}}\text{I-IgG} \) was compared to \( {^{131}}\text{I-HSA} \). To elucidate the role of the radio­nuclide and corresponding labeling method, IgG was labeled with \( {^{111}}\text{In} \), \( {^{123}}\text{I} \) and \( {^{99m}}\text{Tc} \) and HSA was radiolabeled with \( {^{111}}\text{In} \) and \( {^{131}}\text{I} \).

MATERIALS AND METHODS

Radiopharmaceuticals

The biodistribution and kinetics of six radiolabeled proteins were studied.

- **Indium-111-IgG.** (IgG: Sandoglobulin, Sandoz AG, Nuern­berg, FRG). Diethylenetriaminepentaacetic bicyclic anhydride (bicyclic DTPA) was conjugated to the protein according to Hnatowich and colleagues (9). The purified DTPA-conjugated protein was diluted to 2 mg/ml with 0.15 M acetate (pH = 6.5) and sterilized by membrane filtration. Aliquots of 0.5 ml of the conjugate were radiolabeled with \( {^{111}}\text{In} \) (Indium chloride, Amersham International Ltd., Buckinghamshire, UK) via citrate transchelation. The radiochemical purity of all radiopharmaceuti­cals was determined by instant thin-layer chromatography (ITLC) on Gelman ITLC-SG strips (Gelman Laboratories, Ann Arbor, MI) with 0.1 M citrate (pH = 5) as solvent. Labeling effi­ciency was checked batchwise by HPLC on an \( {^{125}}\text{I} \) size exclusion column (Waters-Millipore) with a 0.1 M acetate buffer (pH = 6) as solvent. A dose of 10 \( \mu \)g labeled with 2 MBq \( {^{111}}\text{In} \) was injected intravenously.

- **Iodine-123-IgG.** (IgG: Sandoglobulin, Sandoz AG, Nuern­berg, FRG). IgG was labeled with \( {^{123}}\text{I} \) by means of the Iodo-gen method (Iodo-gen, 28600, Pierce, Rockford, IL) (10). Unbound iodine was removed with Sephadex 25. A dose of 10 \( \mu \)g IgG labeled with 3 MBq \( {^{123}}\text{I} \) was injected intravenously.

- **Technetium-99m-IgG.** Kits for labeling IgG with \( {^{99m}}\text{Tc} \) (Technescan-HIG) were kindly provided by Mallinkrodt Diagnostica Holland, Petten, The Netherlands. A kit, containing 1 mg of 2-­iminothiolane-derivatized IgG (obtained from the Central Labo­ratory of the Bloodtransfusion Service of The Netherlands Red Cross, Amsterdam, The Netherlands) and stannous chloride, was radiolabeled with 500 MBq \( {^{99m}}\text{Tc} \) eluate. In vitro stability was tested by HPLC analysis after in-vitro storage of \( {^{99m}}\text{Tc-IgG} \) at 37°C for 24 hr. A dose of 10 \( \mu \)g, labeled with 4 MBq \( {^{99m}}\text{Tc} \), was injected intravenously.

- **Indium-111-HSA.** (Human Albumin 20%, Central Laboratory of the Bloodtransfusion Service of The Netherlands Red Cross, Amsterdam, The Netherlands). DTPA-conjugation and \( {^{111}}\text{In} \) labeling were accomplished as described above. A dose of 10 \( \mu \)g, labeled with 2 MBq \( {^{111}}\text{In} \), was injected intravenously.

- **Indium-131-HSA.** (Medenix Diagnostics, Fleurus, Belgium). The radiopharmaceutical was obtained commercially. The protein bound \( {^{131}}\text{I} \) activity was 96%. A dose of 40 \( \mu \)g labeled with 0.5 MBq \( {^{131}}\text{I} \) was administered intravenously.

- **Indium-111-IgA.** (Human IgA, 11010, Sigma Chemical Com­pany, St. Louis, MO). DTPA-conjugation and \( {^{111}}\text{In} \) labeling were...
accomplished as described above. IgA activity was determined before and after conjugation with DTPA by immunoelectrophoresis. Non-IgA bound \(^{111}\text{In}\) was removed with Sephadex PD 10. A dose of 10 \(\mu\)g, labeled with 2 MBq \(^{111}\text{In}\), was injected intravenously.

**Animals and Study Design**

A calf muscle abscess was induced in young, male, randomly bred Wistar rats (weight 200-220 g) after ether anesthesia with approximately \(2 \times 10^8\) colony-forming units of \textit{Staphylococcus aureus} in 0.1 ml 50:50% suspension of autologous blood and normal saline. The animals were randomly divided in groups. Twenty-four hours after the inoculation of \textit{Staphylococcus aureus} in the muscle, when swelling of the muscle was apparent, the respective radiopharmaceuticals were injected in the tail vein.

To collect tissues, rats were killed with 30 mg intraperitoneally injected phenobarbital, followed by cervical dislocation at 2, 6, 24, and 48 hr after injection. For \(^{99m}\text{Tc}-\text{IgG}\), tissues were obtained up to 24 hr p.i. Each peptide was evaluated in six animals at each time point. Samples of bone marrow (taken from the right femur), blood, and urine were collected. The infected left calf muscle, the right calf muscle, the liver, the spleen, the kidneys, and the right femur were collected and blotted dry. The activity in the tissues and samples was measured in a shielded well-type gamma counter. The excreted activity in feces and urine of each peptide at each time point between injection of the radiopharmaceutical and killing of the animals was also measured in the well counter. To correct for radioactive decay and permit calculation of the uptake of the radiopharmaceuticals in each organ as a fraction of the injected dose, aliquots of the respective doses were counted simultaneously. The measured activity in tissues and samples was expressed as percentage of injected dose per gram (%ID/g) and the excreted activity per animal as percentage of total dose administered.

For calculation of background activity, we used a combination of normal muscle activity and blood activity. Since the blood volume of rats approximates 60 ml per kg bodyweight (6%), 94% \(\times\) the activity per gram in muscle plus 6% \(\times\) the activity per gram in blood was adopted as the value for background activity (11). Abscess-to-background ratios were calculated.

**Statistical Analysis.**

All mean values are given ± s.e.m. Statistical analysis was performed using Tukey's analysis of variance. The level of significance was set at 0.05.

**RESULTS**

**Labeling Efficiency and Characterization of Proteins**

Conjugation of DTPA to proteins resulted in two to three DTPA ligands per protein molecule. For \(^{111}\text{In}-\text{IgG}\), labeling efficiency was 95%. ITLC and HPLC analysis results were similar. In patients, all \(^{111}\text{In}\) activity in blood samples was still protein-bound at 48 hr p.i. No cell binding of \(^{111}\text{In}\) could be detected.

For \(^{123}\text{I}-\text{IgG}\), radiochemical purity was 98% after removing unbound iodine. For \(^{99m}\text{Tc}-\text{IgG}\), labeling efficiency was 99%. HPLC analysis showed that approximately 75% of the original \(^{99m}\text{Tc}\) activity was still protein-bound after 24 hr in vitro.

For \(^{111}\text{In}-\text{HSA}\) and \(^{131}\text{I}-\text{HSA}\), the protein bound activity was 99% and 96%, respectively. IgA activity after conjugation with DTPA was 89% of the original activity. Indium-111-IgA labeling efficiency was 96% after purification.

**Role of Protein**

Comparison of \(^{111}\text{In}-\text{IgG}\), \(^{111}\text{In}-\text{HSA}\) and \(^{111}\text{In}-\text{IgA}\). Figure 1 shows that all radiopharmaceuticals had a higher accumulation in the infectious focus than in the contralateral normal muscle at all sampling times. Indium-111-IgG uptake in the abscesses remained at a constant, high level. Although not significant, both \(^{111}\text{In}-\text{HSA}\) and \(^{111}\text{In}-\text{IgA}\) showed a tendency to decrease over time. The uptake of \(^{111}\text{In}-\text{IgG}\) and \(^{111}\text{In}-\text{HSA}\) in the abscess revealed no significant differences. Indium-111-IgA uptake was significantly lower at all points in time (\(p < 0.05\)). However, abscess-to-background ratios of all \(^{111}\text{In}\)-labeled proteins (Fig. 2) did not differ significantly.

![FIGURE 1. Activity uptake in abscess and muscle, expressed as %dose/g (error bars indicate s.e.m.).](image-url)
After 6 hr, \textsuperscript{\textit{111}}In-IgG cleared significantly slower from the blood than \textsuperscript{\textit{111}}In-HSA, \textit{p} < 0.05 (Fig. 3). The blood concentration of \textsuperscript{\textit{111}}In-IgA was significantly lower than that of \textsuperscript{\textit{111}}In-IgG and \textsuperscript{\textit{111}}In-HSA at each time point (\textit{p} < 0.05).

Table 1 and Figure 4 show the distribution in various organs. Indium-\textsuperscript{111}-IgA showed marked accumulation in the kidneys, liver and spleen. Indium-\textsuperscript{111}-IgG and \textsuperscript{\textit{111}}In-HSA uptake in the liver and the spleen did not differ significantly. However, renal uptake of \textsuperscript{\textit{111}}In-IgG was significantly higher compared to \textsuperscript{\textit{111}}In-HSA (\textit{p} < 0.05).

Excretion of \textsuperscript{\textit{111}}In-IgG and \textsuperscript{\textit{111}}In-HSA were also similar (Fig. 5). Indium-\textsuperscript{111}-IgA excretion was slightly higher.

Comparison of \textsuperscript{\textit{123}}I-IgG and \textsuperscript{\textit{131}}I-HSA. At 24 and 48 hr, both \textsuperscript{\textit{123}}I-IgG and \textsuperscript{\textit{131}}I-HSA revealed significant washout from the abscess, \textit{p} < 0.05 (Fig. 1). The absolute abscess uptake of \textsuperscript{\textit{123}}I-IgG was significantly higher than the corresponding uptake of \textsuperscript{\textit{131}}I-HSA at 24 and 48 hr, since the washout of \textsuperscript{\textit{123}}I-IgG was slower than that of \textsuperscript{\textit{131}}I-HSA (\textit{p} < 0.05). This also applied for the absolute muscle uptake. For this reason, abscess-to-background ratios of the two iodinated proteins did not differ (Fig. 2).
TABLE 1

Biodistribution, Expressed as Percentage of Administered Dose per Gram (Mean Values ± s.e.m.)

<table>
<thead>
<tr>
<th>Organ</th>
<th>Time</th>
<th>$^{111}$In-lgG</th>
<th>$^{123}$I-lgG</th>
<th>$^{99m}$Tc-lgG</th>
<th>$^{111}$In-HSA</th>
<th>$^{131}$I-HSA</th>
<th>$^{111}$In-lgA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Liver</td>
<td>2</td>
<td>1.14 ± 0.03</td>
<td>0.92 ± 0.11</td>
<td>0.88 ± 0.03</td>
<td>1.07 ± 0.16</td>
<td>0.54 ± 0.04</td>
<td>6.25 ± 0.12</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>1.32 ± 0.07</td>
<td>0.66 ± 0.10</td>
<td>0.77 ± 0.07</td>
<td>1.39 ± 0.19</td>
<td>0.42 ± 0.02</td>
<td>6.94 ± 0.34</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>1.37 ± 0.09</td>
<td>0.26 ± 0.02</td>
<td>0.38 ± 0.08</td>
<td>1.29 ± 0.19</td>
<td>0.12 ± 0.00</td>
<td>5.94 ± 0.19</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>1.49 ± 0.04</td>
<td>0.29 ± 0.03</td>
<td>0.47 ± 0.21</td>
<td>0.05 ± 0.01</td>
<td>4.85 ± 0.33</td>
<td></td>
</tr>
<tr>
<td>Spleen</td>
<td>2</td>
<td>1.09 ± 0.04</td>
<td>0.90 ± 0.10</td>
<td>0.88 ± 0.08</td>
<td>1.12 ± 0.13</td>
<td>0.58 ± 0.05</td>
<td>4.77 ± 0.59</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>1.43 ± 0.10</td>
<td>0.72 ± 0.08</td>
<td>0.77 ± 0.08</td>
<td>1.68 ± 0.15</td>
<td>0.44 ± 0.03</td>
<td>3.70 ± 0.36</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>1.61 ± 0.09</td>
<td>0.36 ± 0.02</td>
<td>0.40 ± 0.02</td>
<td>1.90 ± 0.31</td>
<td>0.12 ± 0.01</td>
<td>4.41 ± 0.60</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>1.81 ± 0.11</td>
<td>0.28 ± 0.02</td>
<td>0.41 ± 0.36</td>
<td>0.05 ± 0.00</td>
<td>3.15 ± 0.48</td>
<td></td>
</tr>
<tr>
<td>Kidneys</td>
<td>2</td>
<td>4.18 ± 0.22</td>
<td>1.47 ± 0.14</td>
<td>9.41 ± 0.84</td>
<td>1.57 ± 0.17</td>
<td>0.93 ± 0.09</td>
<td>7.64 ± 0.20</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>5.45 ± 0.49</td>
<td>1.15 ± 0.08</td>
<td>11.10 ± 0.96</td>
<td>1.62 ± 0.15</td>
<td>0.70 ± 0.03</td>
<td>7.88 ± 0.13</td>
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<td></td>
<td>24</td>
<td>5.38 ± 0.46</td>
<td>0.85 ± 0.03</td>
<td>10.15 ± 0.81</td>
<td>1.74 ± 0.18</td>
<td>0.21 ± 0.01</td>
<td>8.27 ± 0.14</td>
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<tr>
<td></td>
<td>48</td>
<td>5.36 ± 0.61</td>
<td>0.45 ± 0.03</td>
<td>10.20 ± 0.78</td>
<td>2.50 ± 0.31</td>
<td>0.09 ± 0.01</td>
<td>7.81 ± 0.25</td>
</tr>
<tr>
<td>Bone marrow</td>
<td>2</td>
<td>1.01 ± 0.10</td>
<td>1.30 ± 0.10</td>
<td>1.27 ± 0.28</td>
<td>1.19 ± 0.07</td>
<td>0.90 ± 0.17</td>
<td>1.14 ± 0.08</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>1.45 ± 0.08</td>
<td>0.99 ± 0.11</td>
<td>0.78 ± 0.09</td>
<td>1.13 ± 0.18</td>
<td>0.68 ± 0.05</td>
<td>0.99 ± 0.06</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>1.40 ± 0.08</td>
<td>0.40 ± 0.03</td>
<td>0.27 ± 0.01</td>
<td>1.24 ± 0.02</td>
<td>0.20 ± 0.05</td>
<td>0.89 ± 0.07</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>1.40 ± 0.08</td>
<td>0.39 ± 0.02</td>
<td>1.07 ± 0.10</td>
<td>0.06 ± 0.01</td>
<td>0.79 ± 0.09</td>
<td></td>
</tr>
<tr>
<td>Bone</td>
<td>2</td>
<td>0.25 ± 0.02</td>
<td>0.27 ± 0.02</td>
<td>0.22 ± 0.03</td>
<td>0.28 ± 0.04</td>
<td>0.24 ± 0.02</td>
<td>0.18 ± 0.02</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>0.23 ± 0.02</td>
<td>0.23 ± 0.02</td>
<td>0.16 ± 0.01</td>
<td>0.37 ± 0.04</td>
<td>0.18 ± 0.03</td>
<td>0.14 ± 0.01</td>
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<tr>
<td></td>
<td>24</td>
<td>0.28 ± 0.02</td>
<td>0.11 ± 0.01</td>
<td>0.08 ± 0.00</td>
<td>0.39 ± 0.05</td>
<td>0.10 ± 0.01</td>
<td>0.16 ± 0.02</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>0.34 ± 0.05</td>
<td>0.10 ± 0.01</td>
<td>0.87 ± 0.09</td>
<td>0.08 ± 0.00</td>
<td>0.11 ± 0.01</td>
<td></td>
</tr>
</tbody>
</table>

The blood clearance and excretion rate of $^{123}$I-lgG were significantly slower than the clearance and excretion rate of $^{131}$I-HSA $p < 0.05$ (Fig. 3 and 5). The accumulation in organs was significantly lower for $^{131}$I-HSA than for $^{123}$I-lgG, $p < 0.05$ (Table 1 and Fig. 4).

Role of Radionuclide

Comparison of $^{111}$In-IgG, $^{123}$I-IgG, and $^{99m}$Tc-IgG. As shown in Figure 1, absolute abscess uptake over time showed different patterns for these labels: $^{111}$In-IgG was relatively high and constant from 6 hr p.i. onwards; $^{123}$I-lgG was also high at 6 hr p.i., but decreased afterwards; $^{99m}$Tc-IgG was relatively low and decreased between 6 and 24 hr p.i. The uptake of $^{111}$In-IgG in the abscess was significantly higher than that of $^{99m}$Tc-IgG at 24 hr ($p < 0.05$). Also, at 48 hr there was more $^{111}$In-IgG than $^{123}$I-lgG activity in the abscess ($p < 0.05$). Abscess uptake of $^{123}$I-lgG was initially significantly higher than that of $^{99m}$Tc-IgG ($p < 0.05$), but did not differ significantly at 24 hr p.i. With regard to the abscess-to-background ratios (Fig. 2), only the higher ratio at 48 hr p.i. of $^{111}$In-IgG compared to the ratio of $^{123}$I-lgG reached a level of significance ($p < 0.05$).

As shown in Figure 3, significant differences could be noted in blood clearance: $^{99m}$Tc-IgG showed the fastest clearance ($p < 0.05$) and $^{123}$I-lgG the slowest ($p < 0.05$).
The accumulation of the three IgG preparations in the kidneys varied significantly: 123I-IgG had the lowest (p < 0.05) and 99mTc-IgG had the highest uptake (p < 0.05) (Table 1 and Figure 4). In liver, spleen, bone and bone marrow, 111In-IgG uptake significantly exceeded 123I-IgG and 99mTc-IgG uptake (p < 0.05). Indium-111-IgG showed stable or increasing activity over time in liver, spleen, kidneys, bone marrow and bone. Iodine-123-IgG activity decreased in these organs, while 99mTc-IgG activity also decreased in these organs with the exception of the kidneys, whose activity remained relatively stable. Typically, the bone marrow uptake exceeded the bone uptake by a factor four to five for all preparations.

As shown in Figure 5, approximately 20% of the administered 111In-IgG was excreted in faeces and urine within 48 hr, excretion of 123I-IgG was higher (approximately 30%). After administration of 99mTc-IgG approximately 60% of the radiopharmaceutical was excreted within 24 hr.

Comparison of 111In-HSA and 131I-HSA. Similar differences as noted between 111In-IgG and 123I-IgG were found between 111In-HSA and 131I-HSA: retention in the abscess, constant abscess to background ratios over time, significant accumulation in organs, and low excretion rate of 111In-HSA versus washout from the abscess, over time decreasing abscess-to-background ratios, low organ uptake, and high excretion rate of 131I-HSA.

DISCUSSION

The similar abscess uptake and abscess to background ratios of 111In-IgG and 111In-HSA at various time points indicate that specific receptor binding of labeled IgG in an infectious focus, if at all present, is not the major factor in accumulation of labeled IgG in such a focus, since HSA lacks this receptor affinity. These findings are in concert with the autoradiographic studies by Morrel et al. and our own experience in neutropenic patients (7,8).

IgA is an immunoglobulin without Fc-γ receptor affinity. On inflammatory cells few Fc-α receptors are present (12). However, target-to-background ratios of 111In-IgA in infections in extremities do not differ from those after 111In-IgG injection, thus confirming that Fc-γ receptor interaction is not a major factor in the accumulation of 111In-IgG in infectious foci. The different uptake in liver, spleen and kidney of 111In-IgG and 111In-IgA revealed that protein is also a major factor in biodistribution. Since hepatocytes of rats are equipped with IgA receptors, the high liver uptake of 111In-IgA in our experiment is not surprising (13). The initially high and persistent organ uptake of 111In-IgA is in accordance with fast clearance from the blood to the organs. Hepatic, renal and splenic uptake of 111In-IgA are so high, that, at least in rats, this agent is not suited for imaging infectious foci in these areas. The differences observed between 111In-IgG and 111In-IgA confirm the results of Fischman et al., who noted that physical chemical differences between 111In-labeled Fab and Fc fragments of IgG might account for differences in abscess localization and biodistribution (14).

More striking differences were observed with regard to the biological behavior of IgG when it was labeled to either 111In, 123I or 99mTc. Indium-111-IgG was retained in the abscess, while 123I-IgG and 99mTc-IgG showed washout. The physiological uptake in organs of 111In-IgG, the radiopharmaceutical thus far most frequently used in patient studies, is relatively high for most organs, compared to 123I-IgG and, except for kidney uptake, 99mTc-IgG.

The relatively slow blood clearance of 123I-IgG might appear disadvantageous in detection of infectious foci, because of persistently high background activity. The most probable explanation for this persisting blood activity is
Biodistribution of Radiolabeled Proteins • Oyen et al

Both the present study and data in the literature stress the relevance of the radiolabel on the dynamic distribution of the protein. In the present study, great similarity was observed between $^{111}$In-IgG and $^{111}$In-HSA. Although washout of $^{123}$I-IgG from the abscess was slower than that of $^{131}$I-HSA, both iodinated proteins showed similar trends with regard to uptake and retention in the abscess and the organs and the abscess-to-background ratios and distribution. In contrast, Calame et al. observed lower abscess-to-background ratios for $^{99m}$Tc-HSA than for HSA-IgG (19). However, their experimental model differed from ours with regard to the animal, the strain of bacteria and amount of bacteria injected.

In conclusion, both the radionuclide and the protein are important in the biodistribution and kinetics of radiolabeled proteins used for detection of infectious foci. For accumulation in infectious foci, the radionuclide appears to be the major determinant. Specific Fcγ receptor binding is probably of minor, if any, importance in the uptake of radiolabeled proteins in infectious foci. Most probably accumulation in these areas is caused by increased vascular permeability initially and retention with time by macrovascular entrapment. The protein defines the blood clearance and the distribution in organs of radiolabeled proteins, thus determining the amount of labeled protein that is available for delivery to an infectious focus.

Given the similarity of $^{111}$In-IgG and $^{111}$In-HSA in this animal model, further studies in humans with $^{111}$In-HSA would be useful. Despite economic and logistic disadvantages, $^{123}$I-IgG also merits further investigation. The present study warrants further studies to develop a radiolabeled protein with persistently high abscess uptake, low background activity, fast blood clearance and low physiological organ uptake (20).

ACKNOWLEDGMENTS

The authors express their gratitude to: Mr. W. Tax, PhD (University Hospital Nijmegen, Department of Internal Medicine, Division of Nephrology) for his advice; Mr. G. Borm, PhD (University of Nijmegen, Department of Medical Statistics) for performing the statistical analysis; Mr. Gerrie Grutters and Mr. Emile Koenders and Mrs. Maijo van de Ven (University Hospital Nijmegen, Central Animal Laboratory) and Mr. Emile Koenders and Mrs. Marjo van de Ven (University Hospital Nijmegen, Department of Nuclear Medicine) for technical assistance.

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4. Oyen WJG, Claessens RAMJ, van der Meer JWM, Corstens FHM. Detection of subacute infectious foci with indium-111-labeled autologous leukocyte scintigraphy.
The imaging of occult infection is an important area of nuclear medicine. Vehicles for abscess localization have ranged from $^{67}$Ga-citrate to radiolabeled leukocytes to radiolabeled immunoglobulin G (IgG) of current interest. Although $^{111}$In labeled polyclonal IgG is probably the most widely cited protein being evaluated for focal infection scintigraphy, comparison with gallium-67-citrate and technetium-99m-labeled albumin (9) is essential for imaging focal sites of infection. This study was well-conceived and designed to determine the role of protein carrier and radiolabel. However, an accurate interpretation of the role of the protein assumes the radiolabel serves as a radiotracer. Furthermore, an interpretation of the role of the radiolabel requires an analysis of its pharmacokinetics of its radioactive metabolites. Once these properties dictate the biodistribution of radioactively, it is instructive to briefly review relevant factors such as attachment stability, metabolic fate, and route of excretion characteristic of radioisotopes, $^{111}$In and $^{99m}$Tc as used in this study.

IODINE AS RADIOTRACER

Radioiodine isotope continue to be the most widely used protein radio-labels: $^{131}$I for imaging, and $^{125}$I and $^{131}$I for preclinical studies with their convenient longer half-lives and ready availability. The "easy" direct radiiodination approach in which the radioiodine is added to the activated ortho position of tyrosine is most often used, as was done in the Oyen et al. study (9). Label stability is usually sufficient to follow proteins in circulation or bound to cell surfaces. Once internalized by cells, however, catabolism releases peptide fragments or free amino acids with further metabolic processing ultimately releasing radioiodide ($^{131}$I). Deiodination may occur rapidly as in the example of the T-101 antibody in which imaging of cutaneous T-cell lymphoma is virtually precluded by rapid loss of radioactivity from tumor cells (11). Metabolically stabilized ligand chemistry has been developed which substab-

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**EDITORIAL**

Targeted Proteins for Diagnostic Imaging: Does Chemistry Make a Difference?

The image of occult infection is an important area of nuclear medicine. Vehicles for abscess localization have ranged from $^{67}$Ga-citrate to radiolabeled leukocytes to radiolabeled immunoglobulin G (IgG) of current interest. Although $^{111}$In labeled polyclonal IgG is probably the most widely cited protein being evaluated for focal infection scintigraphy, comparison with gallium-67-citrate and technetium-99m-labeled albumin (9) is essential for imaging focal sites of infection. This study was well-conceived and designed to determine the role of protein carrier and radiolabel. However, an accurate interpretation of the role of the protein assumes the radiolabel serves as a radiotracer. Furthermore, an interpretation of the role of the radiolabel requires an analysis of its pharmacokinetics of its radioactive metabolites. Once these properties dictate the biodistribution of radioactively, it is instructive to briefly review relevant factors such as attachment stability, metabolic fate, and route of excretion characteristic of radioisotopes, $^{111}$In and $^{99m}$Tc as used in this study.

IODINE AS RADIOTRACER

Radioiodine isotopes continue to be the most widely used protein radio-labels: $^{131}$I for imaging, and $^{125}$I and $^{131}$I for preclinical studies with their convenient longer half-lives and ready availability. The "easy" direct radiiodination approach in which the radioiodine is added to the activated ortho position of tyrosine is most often used, as was done in the Oyen et al. study (9). Label stability is usually sufficient to follow proteins in circulation or bound to cell surfaces. Once internalized by cells, however, catabolism releases peptide fragments or free amino acids with further metabolic processing ultimately releasing radioiodide ($^{131}$I). Deiodination may occur rapidly as in the example of the T-101 antibody in which imaging of cutaneous T-cell lymphoma is virtually precluded by rapid loss of radioactivity from tumor cells (11). Metabolically stabilized ligand chemistry has been developed which substab-

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